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THE MICROSCOPY OF DRINKING WATER

BY

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FOURTH EDITION

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DEDICATED
TO
MY FATHER AND MOTHER

PREFACE TO THE FOURTH EDITION

The three editions through which this book has passed attest in a measure to its usefulness as a textbook for students in sanitary engineering and sanitary chemistry and to the assistance it has given the water analyst and the water works engineer in solving the problems that have to do with the microscopic organisms of drinking water.

In bringing to a close his preface to the third edition the author expressed his conviction that the micrology of water was going to play an increasingly important part in the science of sanitation. He elaborated on this statement as follows: "The demand for clean water is growing. Popular standards of purity are rising. Our cities need water of such quality that the people not only can drink it with safety, but will drink it with pleasure. 'Safety first' is as good a motto for the water supply service as it is for the railroad service, but safe water that is not also clean loses, psychologically, much of its value."

A review of the technical literature that has appeared since the author thus expressed his conviction shows his prognostication to be true. Money and effort have been and continue to be spent in increasing amounts for the purpose of making water palatable as well as safe. The control of algæ by scientific design and operation of both reservoirs and purification works has advanced greatly. The revisers believe, however, that a wider and deeper study of the practical aspects of microscopic aquatic life and of limnology and rheology can be made to yield yet greater fruitage. It is with this in mind that they have attempted a revision and expansion of the previous work, and sought to make available some of the knowledge acquired in recent years.

The nearly thirty years that have elapsed since the appearance of the book have been marked by an astounding amount of work both in this country and abroad upon the study of microscopic organisms in water. Their systematic relationships, their food requirements and their response to many natural phenomena have been more closely defined. Biologists, chemists, and engineers have all taken a share in this advance of the composite science of aquatic microscopy. Their contributions have made necessary a rearrangement and expansion of Part I which deals with the environment and distribution of organisms and with their collection and examination and of Part II in which the common denizens of water are classified and described.

Part I, "Applied Microscopy," has been rearranged, rewritten and enlarged. On the surface it seems to bear but little relation to the original work, but this is not so in fact. The material has merely been brought together in what appeared to be a more serviceable form, both from the standpoint of teaching and that of practical use.

Chapter I has been prepared with a view of presenting a survey of the development and scope of microscopical science and of the distribution of microscopic organisms in natural waters. Terminology and systematic position of the microscopic organisms have been more adequately treated than in former editions.

Chapter II, "Microscopic Organisms and Sanitary Water Analysis," is new. It is intended to show the parts and purposes of a sanitary analysis and to point out the relations of the microscopical examination of water to other elements of the analysis. It is supported by Chapter III, "Tastes and Odors in Water Supplies," which has been expanded to include a discussion of chlorination odors and tastes, so important in modern water works practice.

The two chapters on "Collection of Samples" and on "Examination of Samples" contain much new material, such as that dealing with larger aquatic organisms, bottom sediments and the use of the centrifuge. An entire chapter is devoted to "Records of Examination" in order to bring out the comparative usefulness of new and old units of measurement and to indicate statistical methods for presenting and studying the results of examination.

"Limnology" has been separated into three parts and treated in as many chapters under the head of "Physical, Chemical and Biological Conditions." Much that is new in precept and example is included in these chapters, which illustrate the effects of the natural environment upon the microscopic organisms and the reciprocal effects of the organisms upon their surroundings.

The use of rivers as water supplies and the rôle of microscopic organisms in streams has not been stressed in previous editions. Chapter XI, "Rheology," has, therefore, been added and treats of the physical, chemical, and biological conditions of flowing waters after the manner that the chapters on "Limnology" treat of those of quiet waters.

The chapter on the "Self-Purification of Streams" is also new. The increasing pollution of our rivers and the part played by aquatic life in bringing about or aiding the natural purification of water are subjects that have a direct bearing on clean and wholesome water supplies. The forces of self-purification, the zones that mark its extent and the methods of measuring its progress are therefore described.

The chapter on "Control of Algæ" has been expanded to contain

some of the material given under Soil Stripping in the third edition. It includes in addition a large amount of new information on copper sulphate and chlorine treatment gathered from experience in this and other countries. New material in Chapter XIV, "Purification of Water Containing Algæ," includes a consideration of further developments in aërotors and aërotor nozzles, the resistance to filtration of algæ-laden waters and the effect of organisms upon filter runs. Chapter XV, "Microscopic Organisms in Water Conduits," contains new information on pipe growths and their effects upon the hydraulic characteristics of conduits in which they occur.

In spite of the wealth of new knowledge that has been available for the work of this revision, emanating both from the researches of pure science and from the experience and observations of practice, there is still great inspiration to carry on; this comes from contemplation of the many factors as yet unknown in the realm of the microscopy of water. The sudden coming and going of immense numbers of organisms in lakes and reservoirs, the true rôle of microscopic life in the natural purification of polluted waters, the control of troublesome growths, all these and other subjects are only partially understood and urgently call for further fundamental study and empirical formulation.

Part II, "Determinative Microscopy," has also been subjected to study and revision. This section of the book might easily have been enlarged to contain descriptions and plates of many other organisms important to sanitarians, particularly of those forms intimately connected with the debasement of clean water by wastes from cities and industries and with its subsequent natural purification. Such a departure, however, would have unduly added to the size and expense of the book and would have altered the main purpose for which it was written, namely to serve as an elementary guide to the microscopy of water. Also, there has appeared since the last edition a comprehensive book on "Fresh Water Biology," sponsored by Dr. Henry B. Ward and the author, which meets the needs of those who wish to proceed beyond the confines of this work. All are urged to do so.

In order to familiarize the student with advanced methods of systematic classification and identification of organisms and for the purpose of correlating the information contained in this book with that in other more extensive works, keys to the common forms of organisms have been added to those chapters of Part II in which their use was warranted. The keys to the algæ were made quite extensive because the organisms of this group are of particular significance in the study of drinking water.

Chapter XXXII, "Ecological Classification of Microscopic Organisms," constitutes an important addition to Part II and will serve to

broaden its horizon. Here are listed some thousand aquatic organisms, identified as to their preferred environment. The organisms are not given because they are found in drinking water, — many of them are not, — but they are here assembled because they are potent factors in the natural purification of water, the study of which touches intimately upon many of the problems connected with drinking water supplies. A new and added significance has been attached in recent years to the conservation of our streams; even greater attention will be focused upon the subject in the future. It is hoped that Chapter XXXII together with Chapter XII will stimulate interest in the biological aspects of self-purification and that it will serve as a stepping stone to a more accurate and exact knowledge of the ecology of aquatic organisms than exists to-day, particularly as regards the organisms of our own waters.

References to the literature have been brought up to date. Many of the older references have been omitted to make room for newer ones of equal or greater importance. The references have been classified according to subject matter by appending them to the various chapters in which they are referred to or which they otherwise concern.

The Glossary has been expanded to include all but the most familiar scientific terms used in this book.

With all the pleasure that accompanies the completion of this revision there comes the sorrowful duty of recording the passing of the author of this book. His loss has been to the revisers, as to so many others, that of a great teacher, an inspiring colleague, a wise counselor and a dear friend. One of his planned tasks was a thorough revision of this — his first — book, the scope of which he desired to enlarge by the addition of a chapter on stream purification and by the inclusion of some of the newer aspects of microscopy. It is the hope of the revisers that their close association with the author for many years before his death has enabled them to carry out his plans in the same spirit, if not to the same degree of excellence, with which he would have accomplished the task.

Throughout the course of recent months the work of the revisers has been greatly lightened and expedited by valued assistance from many persons. The section in Part II dealing with the algæ and fungi was carefully reviewed and revised by Prof. C. W. Dodge, that on the protozoa by Dr. J. A. Dawson, on the rotifera by Mr. G. L. Walls, on the crustacea by Mr. L. A. Brown, all of Harvard University. Much of the newer literature was reviewed by Mr. T. F. Hatch, Instructor in Sanitary Engineering. Valuable suggestions relating to the revision were received from Dr. Frank E. Hale of Mt. Prospect Laboratory, Mr. W. C. Purdy of the U. S. Public Health Service, Dr. Chancey Juday

of the University of Wisconsin, Mr. Robert E. Richardson and Dr. A. M. Buswell of Illinois, and many others. To all the revisers owe a debt of gratitude for their generous responses. They also gratefully acknowledge the services of their secretary, Miss Evelyn Barnes, for her painstaking care in preparing the manuscript.

GORDON MASKEW FAIR.
MELVILLE CONLEY WHIPPLE.

CAMBRIDGE, *March* 1, 1927.

PREFACE TO THE FIRST EDITION

This book has a twofold purpose. It is intended primarily to serve as a guide to the water analyst and the water works engineer, describing the methods of microscopical examination, assisting in the identification of the common microscopic organisms found in drinking water and interpreting the results in the light of environmental studies. Its second purpose is to stimulate a greater interest in the study of microscopic aquatic life and general limnology from the practical and economic standpoint.

The work is elementary in character. Principles are stated and briefly illustrated, but no attempt is made to present even a summary of the great mass of data that has accumulated upon the subject during the last decade. The illustrations have been drawn largely from biological researches made at the laboratory of the Boston Water Works and from the reports of the Massachusetts State Board of Health. In considering them one should remember that the environmental conditions of the Massachusetts water supplies are not universal, and that every water supply must be studied from the standpoint of its own surroundings. As far as the microscopic organisms are concerned, however, the troubles that they have caused in Massachusetts may be considered as typical of those experienced elsewhere.

The descriptions of the organisms in Part II are necessarily brief and limited in number. The organisms chosen for description are those that are most common in the water supplies of New England, and those that best illustrate the most important groups of microscopic animals and plants. In many cases whole families and even orders have been omitted, and some readers will doubtless look in vain for organisms that to them seem important. The omissions have been made advisedly and with the purpose of bringing the field of microscopic aquatic life within the scope of a practical and elementary survey. For the same reason the descriptions stop at the genus and no attempt has been made to describe species and varieties. Notwithstanding this it is believed that the illustrations and descriptions are complete enough to enable the general reader to obtain a true conception of the nature of the microscopic life in drinking water and to appreciate its practical importance. To the student they must serve as a skeleton outline upon which to base more detailed study.

The illustrations, for the greater part, have been drawn from living specimens or from photo-micrographs of living specimens, but some of them have been reproduced from published works of standard authority. Among these may be mentioned: Pelletan and Wolle on the Diatomaceæ; Wolle, Rabenhorst, and Cooke on the Chlorophyceæ and Cyano-phyceæ; Zopf on the Fungi; Leidy, Bütschli, and Kent on the Protozoa; Hudson and Goss on the Rotifera; Baird and Herrick on the Crustacea; Lankester on the Bryozoa; Potts on the Spongidiæ; and Griffith and Henry on miscellaneous organisms.

This book has been prepared during the leisure moments of a busy year. Its completion has been made possible by the kind assistance of my present and former associates in the laboratories of the Boston and Brooklyn water supply departments and of other esteemed friends, to all of whom I tender my sincere thanks. I desire also to acknowledge the valuable assistance of my wife, Mary R. Whipple, in revising the manuscript and correcting the proof. To many others I am indebted indirectly, and among them I cannot refrain from mentioning the names of Prof. W. T. Sedgwick of the Massachusetts Institute of Technology; Mr. Geo. W. Rafter, C.E., of Rochester, N. Y.; and Mr. Desmond FitzGerald, C.E., formerly Superintendent of the Boston Water Works and now Engineer of the Sudbury Department of the Metropolitan Water Works. To Prof. Sedgwick and Mr. Rafter water analysts are indebted for the most satisfactory practical method of microscopical examination of drinking water yet devised, and Mr. FitzGerald will be remembered not only as an eminent engineer but as the founder and patron of the first municipal laboratory for biological water analysis in this country.

GEORGE CHANDLER WHIPPLE.

NEW YORK, *January*, 1899.

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THE MICROSCOPY OF DRINKING WATER

PART I

APPLIED MICROSCOPY

CHAPTER I

MICROSCOPIC ORGANISMS IN DRINKING WATER

The microscopy of drinking water is an applied science which draws its basic information from the larger subject of fresh-water biology. The latter deals with all the forms and phenomena of life found in fresh water whether visible to the naked eye or rendered visible only by the use of the compound microscope. It treats of fish and insects, worms and large plants, microscopic organisms and bacteria, in short, of all the forms of life, large and small, that swarm in the waters upon the earth. To the biologist the study of fresh-water life presents one of the most delightful fields of investigation, for in it he finds the most primitive forms of life as well as highly organized multicellular structures. To the geologist it furnishes information concerning vital changes in the earth's development, especially when the environment is considered as well as the organisms themselves. To the chemist it offers a field of study in which minute quantities of the chemical elements as determined by quantitative analysis are made to react and interact to produce remarkable chemical changes. To the physicist it propounds questions of mechanics, optics, and heat that in part still await satisfactory answers. To the sanitarian, finally, it explains the behavior of water and its impurities with relation to human welfare in the light of physics, chemistry, and biology.

Although this book is concerned primarily with the microscopic life in water, excluding the bacteria, it is impossible to consider this group of organisms without touching in one way or another upon the bacteria and upon the larger members of the aquatic community. Similarly, although the subject matter of the book is "drinking water" it will be

found that in order to follow the course that many waters take before reaching the consumer as "drinking water" it will become necessary to digress into other fields such as the self-purification of streams — a subject more closely associated with sewage disposal but very significant in water supply.

Air, water, food, light, and heat are the five essentials for human existence. About them is written the history of mankind. Fundamental as they are, however, how little thought does the average citizen of to-day give to them! Take drinking water, for example: how scant is the information of the great mass of people about it! An abundant supply of wholesome water is taken for granted in all civilized communities of the world. Only calamities, such as a typhoid fever epidemic, a serious water shortage, and a foul odor or taste, bring the lack of clean and sufficient water to the attention of the individual. Then, suddenly, the problem looms large and he asks the questions that every man should ask about those things which are essential to his continued harmonious existence.

That the microscopic life of water in particular is closely associated with the comfort and well-being of man will appear from a study of the many phases of activity involved in "the microscopy of drinking water."

Terminology.* — The term "microscopic organisms" is unfortunately of itself somewhat obscure. By convention it describes in hydrobiology those minute organisms (excepting the bacteria), invisible to the naked eye, which inhabit streams and ditches, pools, ponds and lakes, waters above the ground and in the ground. The basis of this convention is explained in the following scheme suggested by Professor Sedgwick.

MICROÖRGANISMS.

Organisms, either plants or animals, invisible or barely visible to the naked eye.

Microscopic Organisms. (Plankton.)

Not requiring special culture.
Easily studied with the microscope.
Microscopic in size, or slightly larger.
Plants or animals.

Bacterial Organisms.†

Requiring special cultures.
Difficultly studied with the microscope.
Microscopic or sub-microscopic in size.
Plants.

* Attention is called to the glossary at the end of this book. Here most of the scientific terms used in water microscopy are explained.

† The bacteria are not considered in this volume. The reader is referred to the numerous works on Bacteriology, and especially to Prescott and Winslow's "Elements of Water Bacteriology."

This subdivision is convenient for the sanitarian as well as for the biologist, because the two classes of organisms affect drinking water in different ways. With certain reservations it may be said that bacteria make drinking water unsafe, microscopic organisms make it unsavory.

In addition to this general term "microscopic organisms," the minute living things abounding in water are further distinguished by terms descriptive of their mode of occurrence, their size or their nature.

To that assemblage of marine organisms that floats free in water and drifts about with waves and currents, the German oceanographer Hensen gave in 1887 the name *plankton* from the Greek word for "wandering." Plants attached to the shore, and animals that possess strong powers of locomotion, were not considered part of the plankton, but fragments of shore plants, fish eggs, and the like were included. The use of the term has since then been extended to include assemblages of organisms found in the open water of inland lakes and streams. The lake-dwelling organisms in turn have been designated *limnoplankton* (lake plankton) by Hæckel. The free-floating plankton population of rivers has been distinguished by Zacharias as *potamoplankton* (river plankton); the term *rheoplankton* (stream plankton) is probably preferable as including the drifting life in all flowing waters. The passively floating débris that Hensen included in the plankton is now commonly classified as *pseudoplankton*. The individual organisms composing the plankton are known as *planktonts*.*

As opposed to the passively floating plankton, that assemblage of organisms the members of which by their own efforts freely change their location is designated *nekton* from the Greek word for "swimming." The sessile growths found on shore or bottom attached to rocks, pieces of wood, or to the ooze itself are called *benthos* from the Greek word designating the bottom of the sea. The animal forms crawling along shore or bottom surfaces are included in the benthos. In the course of time these specifications have become somewhat neglected and the term "plankton," except in exact technical writing, has been used loosely in a sense practically synonymous with the term "microscopic organisms" of the sanitary biologist.

The regions from which microscopic organisms are gathered are sometimes described as (1) the *limnetic region* or open water, (2) the *littoral region* or shore, (3) the *benthic region* or bottom.

The different methods of collecting plankton have given rise to terms descriptive of the size of the organisms caught. Thus we have the *net plankton* or *mesoplankton*, meaning those organisms that are retained by the plankton net, while the organisms that escape through its meshes

* Recent German writers prefer the term *plankter* as philologically more correct.

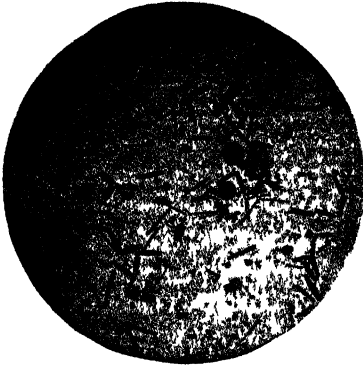
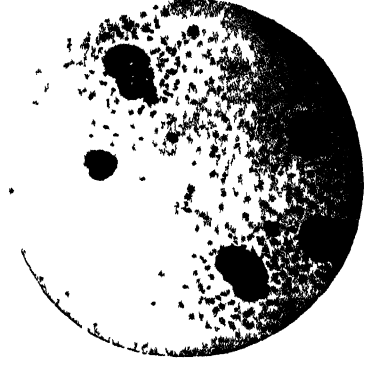
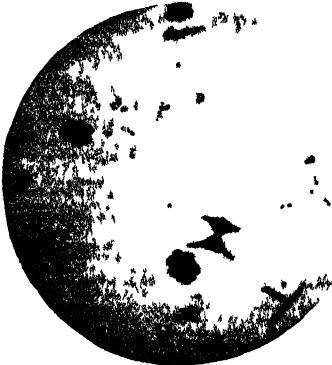
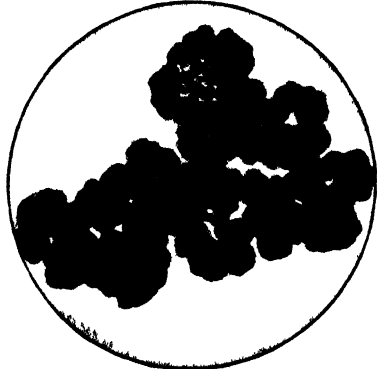
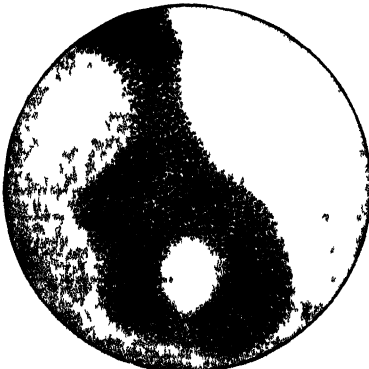
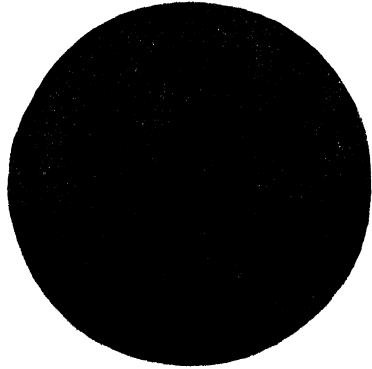
Diatoms. $\times 75$.Cœlosphærium $\times 100$.Pandorina and Staurastrum. $\times 100$.Microcystis $\times 100$ Clathrocystis. $\times 100$.Stigeoclonium. $\times 100$.

PLATE A.

Photomicrographs of Microscopic Organisms Found in Water.
(By John W. M. Bunker.)

have been called *nannoplankton* (dwarf plankton) or *microplankton*. A third group, the *macroplankton*, is visible to the naked eye.

Both the plant and animal kingdoms are represented in the plankton. The members of the former are distinguished by the term *phytoplankton* (plant plankton), and those of the latter by the term *zooplankton* (animal plankton). These in turn are divisions of the *hydrophytes* and *hydrozoa*, respectively, terms that describe all aquatic plant and animal life. The term *algæ* (singular, *alga* = seaweed) in its common usage is synonymous with the term "phytoplankton."

The sudden appearance in lakes and ponds of a surface scum composed of enormous numbers of *algæ* or similar organisms is known as the phenomenon of *water bloom*, which is also variously called *working* or *blooming of the lakes*, *breaking of the meres*, *flos aquæ*, and *Wasserblüte*. Blue-green *algæ* are commonly responsible for this phenomenon.

Position of Microscopic Organisms in the Scale of Life. — The following outlines of fresh-water plant and animal groups suggest the position of the microscopic organisms in the scale of life. Read from top to bottom, these outlines proceed, broadly speaking, from the higher to the lower forms of living things. They are expanded in part to show a classification of the various groups of organisms that are of importance in sanitary microscopy and in sanitary biology in general. Matters of classification and nomenclature are further considered in Chapter XVI, Part II, and the characteristics of the various groups, genera, and species are explained in other chapters dealing with determinative microscopy. The general appearance of some of the various groups of microscopic organisms is indicated in the photomicrographs assembled in Plate A. The different genera are further shown in photographic and colored plates near the end of the book. Appreciation of the relation of microscopic life to its environment and *vice versa* presupposes a certain familiarity with the organisms themselves, and a study of the second part of this book must go hand in hand with that of the first.

OUTLINE OF PLANT GROUPS

SPERMATOPHYTA or *seed plants*.

PTERIDOPHYTA or *ferns*.

LYCOPSIDA or *club mosses*.

BRYOPHYTA or *mosses and liver worts*.

THALLOPHYTA or *thallus plants*.

Fungi.

Characeæ.

Algæ.Chlorophyceæ or *green algæ*.Xanthophyceæ or *yellow-green algæ*.Diatomaceæ or *diatoms*.Phæophyceæ or *brown algæ*.Rhodophyceæ or *red algæ*.**SCHIZOPHYTA or fission plants.**Schizophyceæ* or *fission algæ*, Cyanophyceæ or *blue-green algæ*.Schizomycetes* or *fission fungi*, including the bacteria.**OUTLINE OF ANIMAL GROUPS**VERTEBRATA including the Pisces or *fish* and the Amphibia.MOLLUSCA or *mollusks*.BRYOZOA (Polyzoa) or *moss animalcules*.ARTHROPODA including the Insecta, Arachnida or *water spiders*, mites and bears, and the Crustacea.VERMES including the Cœlhelminthes or *segmented worms*, Trochhelminthes† or *trochal worms*, Nematelminthes or *roundworms*, and the Platyhelminthes or *flatworms*.HYDROZOA or *polyps and medusæ*.PORIFERA or *sponges*.PROTOZOA or *single-celled animals*.Sarcodina or *amœboid protozoa*.Mastigophora or *flagellate protozoa*.Infusoria or *ciliate protozoa*.

Sporozoa (exclusively parasitic).

HISTORICAL

The study of microscopic organisms naturally dates back to the period of scientific investigation following the invention in 1590 of the compound microscope by Hans Jansen and his son Zacharias of Middelburg, Holland. The fact, however, that these minute organisms sometimes develop in vast numbers in water and produce the phenomenon of water bloom provides us with earlier references to the subject or speculation upon it. Thus we find in the Book of Exodus (7, 20 to 25) the account of one of the seven plagues of Egypt in which "all the waters that were in the river were turned to blood, and the fish that

* The Schizophyceæ or Cyanophyceæ are more commonly classed with the algæ and the Schizomycetes with the fungi.

† The Rotifera or *wheel animalcules* are the largest group of the Trochhelminthes.

were in the river died; and the river became foul, and the Egyptians could not drink water from the river." How probable it is that we have here a picture of a "water calamity" induced by the sudden growth of microscopic organisms, notably those blue-green algæ that impart to water, when viewed by reflected light, a deep red color!

Systematic Microscopy. — The true nature and structure of microscopic life in water remained obscure until the end of the seventeenth century. Part of this life has now been traced back by means of fossil remains to the earliest geologic ages. The first to describe microscopic organisms in water was Anton von Leeuwenhoek, who is, as far as sanitary science is concerned, the outstanding scientific amateur. Following Leeuwenhoek's discovery a type of scientific buffoonery seems to have advanced the study of microscopic organisms. The painter Roesel von Rosenhof, for example, wrote a treatise entitled "Insect Divertisements" (*Insektenbelustigungen*) in which he described for the entertainment of his readers many of the common forms of plankton. The mobile or animal organisms naturally attracted most attention, and to them was ascribed, in the beginning, the power to spread disease and pestilence. In the algæ the first observers saw developmental or degenerative stages of the higher plants.

About one hundred years elapsed between the discovery of microscopic organisms and their first classification by a competent scientist. It was in 1786 that the Danish biologist Otto Friedrich Müller published his great work "Animalcula infusoria fluviatilia et marina." The term "infusoria" in these early days was applied to all minute plants and animals and originated in the fact that infusions of decaying organic matter were found to be particularly rich in microscopic life. The term is now restricted to one group of protozoa. In 1838 Ehrenberg made his classical contribution to the study of microscopic organisms and his name will be found connected with many common forms of microscopic life. Since then systematic biological studies of these lowest types of life have progressed by leaps and bounds. The workers in this field have been many. Suffice it merely to mention a few outstanding names, such as Kent, Wollé, Stokes, Zacharias, Kofoid, West, Conn, Tilden, Steuer, Calkins.

Sanitary Microscopy. — Not till the middle of the past century was it recognized that the study of organisms in water had a practical significance. Dr. Hassall of London, England, was the first to call attention to the value of microscopic examinations in the interpretation of drinking-water analyses. His method of procedure is unknown, but in all probability consisted of the examination of a few drops of the sediment collected after allowing the water to stand for some time in a

deep vessel. About the same time Ferdinand Cohn (1853) working on the continent of Europe, wrote his treatise on "Living Organisms in Drinking Water," in which he indicated the correlation between aquatic life and water purity. This he amplified in his subsequent work on *Crenothrix* in well waters.

Beginning with Cohn, microscopic examinations received greater attention in sanitary studies of water. We soon have Professor Hirt expressing, in his book "On the Principles and Methods of Microscopic Analysis of Water" (1879), the relationship of aquatic microscopy to water analysis as follows:

The microscopic examination of water has as its first purpose to control or verify the chemical analysis, as its second to round it out and enlarge it. When the chemist tells us that there is such and such a quantity of "organic matter" in the water analyzed we must of necessity be satisfied with his report even though we do not know what the character of this matter is; the microscopist, however, is in a position to inform us as to its nature especially as regards its morphology.

In spite of these early advances, microscopic analyses were undertaken generally only at times when growths of organisms, explosive in the suddenness and extent of their development, precipitated what have been termed "water calamities," such as the much-cited calamities of Berlin and Rotterdam.

To the Massachusetts State Board of Health belongs the credit of having begun as early as 1887 a systematic examination of all the water supplies of the state. This has been maintained ever since. Two years later the State Board of Connecticut began a similar but less extensive survey. In 1889 the Water Board of the City of Boston established at Chestnut Hill Reservoir a laboratory for the systematic study of the biological character of the various sources of water supply. For the first eight years of its existence the laboratory was conducted by the author under the general direction of Desmond FitzGerald. In 1897 Mt. Prospect Laboratory, connected at first with the Department of Water Supply of Brooklyn, N. Y., and shortly afterwards with all the water supplies of Greater New York, was established. It was devoted to general water analysis, and microscopical examinations formed an important part of the routine and special work. From 1897 to 1904 Mt. Prospect Laboratory was under the direction of the author; from 1904 to 1913 it was in charge of Dr. D. D. Jackson. Since then it has been headed by Dr. Frank E. Hale. This laboratory has produced outstanding work in the control of algæ in water supplies.

Similar biological work has since been undertaken by boards of health and water departments in all parts of the world. Sir Alexander

Houston's work with the London Metropolitan Water Board is particularly noteworthy.

Some of the earliest publications on sanitary microscopy following Hirt's book are Rafter's "The Microscopic Examination of Potable Water" (1892), Mez's "Microscopic Water Analysis" (1898), and the first edition of "The Microscopy of Drinking Water" (1899).

A new note was introduced into the field of sanitary microscopy by the work of Moore and Kellerman in 1905 when they used copper sulphate to eradicate growths of microorganisms from water. More recently, superchlorination for the destruction of microorganisms together with the odors or tastes produced by them has attracted attention.

Microscopy in Limnology and Rheology. — In 1867 another Danish biologist by the name of Müller (Peter Erasmus) observed in Swiss lakes the presence of lower crustacea. Up to his time the water in clear lakes was believed to be free from microscopic life. Müller's studies laid the foundation for limnological research, i.e., the study of lakes. He aroused in Switzerland the interest of Professor F. A. Forel whose work on Lake Geneva, described in "Le Léman," became epoch-making. It resulted in the establishment of a Limnological Commission in Switzerland, under the direction of which many valuable lines of physical and biological research were undertaken. An International Commission followed in 1890. From this time increased attention has been given to the biology of ponds and lakes. In Germany a biological station was established by Zacharias at Lake Plön in 1891, and a group of scientists has contributed a long series of important articles to its reports, which have been published annually since 1893. Apstein at Kiel, Germany, Schröter at Zürich, Switzerland, Wesenberg-Lund at Hilleröd, Denmark, and many others made extensive and valuable observations. Biological stations multiplied, and the work extended to France, Italy, Austria, Sweden, Norway, Scotland and other countries.

Special attention should be called to the work of Sir John Murray and his associates in Scotland. The results of their studies are embodied in "Bathymetrical Studies of the Scottish Lakes." The European writings on the subject are now very voluminous. Abstracts of most of the important articles may be found in the "Internationale Revue der Gesamten Hydrobiologie und Hydrographie," a monthly journal.

Investigations similar to the European ones were also carried on in the United States. In 1893 Prof. J. E. Reighard, acting under the direction of the Michigan Fish Commission, made a biological study of Lake St. Clair. This was followed by an examination of Lake Michigan by Prof. Henry B. Ward, and by studies of the crustacea in Lake Mendota by Prof. E. A. Birge, and in Green Lake by Prof. C. Dwight Marsh.

Biological stations were established by a number of western universities on or in the vicinity of the Great Lakes, and on the shores of smaller bodies of water. Much of the work reaches to the present day.

The development of limnology has its counterpart in that of rheology (the study of streams) and in oceanography. These three branches of hydrography have been mutually stimulating and much of the work developed for one has been extended with success to one or both of the others. The rheological studies of the Illinois River system, begun by Kofoid in 1894 and continued up to the present day at first by him and later by Forbes and Richardson, deserve special mention, as do the investigations of the United States Public Health Service of the Potomac, Ohio, and Illinois rivers.

Microscopic Ecology. — One of the most important later developments in the realm of aquatic microscopy has been the information acquired in relation to microscopic ecology, i.e., the study of the mutual relations between microscopic organisms and their environment. This information has been chiefly of importance in the study of the self-purification of water. The important part played by aquatic life in the natural purification of water was sensed by Forel who in 1895 wrote as follows in regard to Lake Geneva:

The water of the lake is inhabited by a large animal and plant population. Animals, plants, and protista develop here abundantly and live here. By the action of the admirable balance of their opposite functions they labor efficaciously to maintain the waters in a beautiful state of transparency and purity. As soon as the inflowing waters carry into the lake too much dissolved organic matter it is assimilated by the chlorophyllaceous plants and decomposed by the microbes of fermentation. The organic debris is eaten by the protozoa, animal cadavers are devoured by the animals or transformed into gases by the putrefactive bacteria, vibria and micrococci; carbonic acid is reduced by algal and diatomaceous vegetation. The presence in the water of animal and plant life is an element that regulates the proportion of organic matter and dissolved gases; it is a gauge of water purity.

On the other hand it is possible to judge in a way the quality of the water by the grouping of the living things in it. The biological associations of swamp waters, putrid waters, sewage and unwholesome waters are known, likewise those of pure water, fresh water, aerated water, wholesome water. To this last group belong the fauna and flora of large lakes and in particular those of Lake Geneva. Among the animals or plants that inhabit it are a considerable number that are absolutely characteristic of waters of good quality. They could not live in polluted water.

The effect of sewage and trade wastes on the fauna and flora of streams had been observed as early as 1873 by Gérardin who reported on the noxious condition arising from the discharge of organic wastes into the river Croult near Paris. Fish and mollusks died; snails, by climbing up on the stems of water weeds, attempted to escape from the foul water; frogs deserted the stream. The water became putrid, and,

attached to one water wheel alone, there were found over 40 pounds of *Beggiatoa* (sulphur bacteria). As a result of these manifestations Gérardin's attention was called to the importance of the oxygen requirements of aquatic organisms in judging the sanitary quality of water.

Ecological classification of aquatic organisms has been undertaken by many authorities, notably by Kolkwitz and Marsson, members of what is now the *Landesanstalt für Wasserhygiene* in Berlin, Germany.

PURPOSE OF MICROSCOPIC EXAMINATION OF WATER

Microscopic examination of water may serve any one or more of the following purposes:

1. To explain the cause of color and turbidity and the presence of objectionable odors and tastes in water and to indicate methods for their removal.
2. To aid in the interpretation of the chemical analysis.
3. To identify the source of a water (in special cases).
4. To explain the clogging of pipes and filters and aid in the design and operation of water works.
5. To indicate pollution by sewage or trade wastes.
6. To indicate the progress of the self-purification of streams.
7. To aid in explaining the mechanism of biological sewage-treatment methods.
8. To aid in the study of the food of fish, shell fish, and other aquatic organisms.

Color, Turbidity, Odors, and Tastes. — Perhaps the most important service that the microscopical examination renders is that of explaining the cause of the odor, taste, color, and turbidity of many waters. A number of the common microscopic organisms produce objectionable odors or tastes in water. Microscopic examination will determine the nature of the offending organisms and indicate the best method of corrective treatment. When sufficiently abundant, microscopic growths have a marked influence on the color of the water. Certain organisms give the water a green color, others impart pink and even deep red tinges to lakes or reservoirs. These are apparent colors and disappear when the organisms are removed by filtration. Death and decay of chlorophyllaceous growths, however, result in the liberation of the cell content of the organisms and increase the true color of the water, i.e., color due to matter in true or in colloidal solution, which is not removed by passage through filter paper. Plankton growths, furthermore, make the water turbid and produce unsightly scums (water bloom). Upon all these matters that affect the æsthetic quality of water, the

microscopical examination is almost the only means of obtaining reliable information. The information acquired will serve to indicate the most suitable method of removing objectionable conditions created by the growth of microscopic organisms.

Interpretation of Chemical Analysis. — The rôle of microscopic organisms as food consumers and food producers results in their bringing about in their environment certain chemical changes that are reflected in the chemical analysis of the water in which the organisms are found. These changes influence to a certain extent the nitrogen content, gases, hardness, and alkalinity of the water. Furthermore, the presence of living organic matter itself affects such determinations as albuminoid and free ammonia and oxygen consumed. The microscopical examination is, therefore, a valuable aid in the interpretation of chemical findings.

Origin of Waters. — The presence of certain microscopic organisms in water sometimes gives a clue as to its origin. In this way the presence of surface water in a well may be detected. When the Chicago Drainage Canal was opened, the presence of Lake Michigan water in the St. Louis water supply was indicated by the finding of a species of diatoms characteristic of Lake Michigan water.

Clogging of Pipes and Filters. — Filters in both rapid and slow sand plants are often found to clog rapidly. This at times is due to growths in the supply reservoirs of microscopic organisms, particularly blue-green algæ which are quite sticky in nature and form a resistant coating on the filters. At other times clogging is caused by the growth of organisms on the filtering medium itself. This happens in sewage-treatment works as well as in water-purification plants. Attached growths of microscopic organisms, furthermore, may cause serious trouble by clogging water pipes.

Microscopical examination will indicate the corrective treatment to be applied to lengthen filter runs and prevent pipe troubles. It will also aid in determining upon the best methods of purifying waters rich in algal life.

Sewage and Trade Waste Pollution. — Microscopical examination cannot show definitely whether sewage or trade waste is present in a water of the kind that would ordinarily be considered for use as a drinking-water supply. It can, however, give evidence which, considered together with the chemical and bacterial examinations, may establish proof of pollution. A microscopical examination of sewage reveals few of the living organisms that are found ordinarily in water. Ciliated protozoa, fungous forms, and miscellaneous objects, such as yeast cells, starch grains, fibers of wood and paper, muscle fibers, epithelial cells, threads of silk, wool, cotton, and linen, insect scales and feather barbs

may be observed. Most of these objects are foreign to unpolluted water, and their presence in a sample of water leads one to suspect its purity.

Furthermore, there are some organisms that live on decaying vegetable matter and, though not found in sewage, are often associated with it in polluted water. Their presence in a sample gives cause for suspicion. Such evidence, however, should be weighed only in connection with an environmental study or sanitary survey and with the complete sanitary analysis.

The effects of sewage and of organic trade wastes are much alike. There are certain trade wastes, however, both organic and mineral, that are toxic to microscopic life. Discharged into water, such wastes will cause destruction of the aquatic flora and fauna; the absence of microscopic organisms will then give evidence of the character of the polluting substances.

Microscopic Organisms and Disease. — The microscopic organisms commonly encountered in drinking water — algæ, fungi, protozoa, rotifera, crustacea, bryozoa, and sponges — are themselves not the cause of disease in man or in the higher animals. They are not parasitic, and they do not thrive in grossly polluted water. Their presence, therefore, does not indicate sewage pollution. An unusually heavy mass occurrence in drinking water of the common microscopic organisms can, however, conceivably produce nausea or digestive disturbances in water users, but water of this character would generally be too offensive to be consumed without treatment. The production of a specific condition of disease traceable to the drinking of water containing the common microscopic organisms has not been recorded.

The so-called water-borne diseases are caused by parasitic bacteria, protozoa, and worms. All of these organisms can be studied under the microscope; but the pathogenic bacteria are too small to be identified by the common methods of microscopic water examination, and they, as well as the pathogenic protozoa and the ova or larvæ of such endoparasites as flukes, hookworms, tapeworms, and roundworms, are seldom present in water in sufficient numbers to be detected by a sanitary microscopic analysis.

Self-purification of Streams. — The progress of the self-purification of streams may be determined by noting the changes along the course of the stream in the character of microscopic growths and other aquatic life. Certain organisms in particular are sensitive indicators (*Leit-organismen*) of the conditions of existence obtaining in their aquatic environment. They are living reagents that react in certain known ways to their surroundings and thus permit the analyst to draw con-

clusions with respect to the quality of the water in which they are found. It is for this use that microscopical ecology is of great importance.

The rôle of microscopic organisms in bringing about the natural purification of waters is second only to that of the bacteria. While we commonly think of self-purification in connection with sewage or trade-waste pollution it is well to bear in mind that the same processes that may slowly bring about the return of a putrid, sewage-sick stream to a clear and wholesome water course are active also in unpolluted waters that nevertheless are unfit or undesirable for consumption on account of their content of organic or other impurities derived from swamps, soil wash, etc. Self-purification is as important in water supply as it is in sewage disposal. Although we commonly speak of self-purification in connection with streams the phenomena of natural purification can be observed in the standing water of lakes, ponds, and reservoirs, as well as in the flowing water of brooks, rivers and canals.

Biology of Sewage Treatment. — Most modern methods of sewage treatment are dependent in one way or another on biological activity. The processes induced in biological sewage treatment are similar to those obtained in the natural purification of water. In the past the flora and fauna of sewage works have been studied only incidentally and no attempt has been made to cultivate such growths of organisms as will best perform the work required to be done. The new trend in sewage disposal is toward a better understanding of the biological balances that must be maintained between different groups of organisms and between the organisms and the liquids undergoing treatment in order that the best results may be obtained in the most economic way. The activities of the biological workmen of sewage purification are being more and more appreciated.

Food Supply of Fish Life. — Microscopic organisms form the basis of the food supply of fish and other aquatic animals. Forbes has summed up this relation by saying, "No plankton, no fish." Some microscopic organisms are themselves eaten by fish. This was shown by Peck in his study of the menhaden. This fish when feeding swims with its mouth open. The water enters the mouth and passes out through the gills which act as a filtering apparatus by which the minute organisms are caught. It was found that the presence or absence of these fish from certain sections of the Massachusetts coast depended upon the abundance of microscopic life in the water, and also that the weight of fish of any particular length depended upon the quantity of this food material at hand.

The relationship between plankton and fish life may be quite complicated. In many cases the fish feed upon crustacea and insect larvæ;

the crustacea prey upon rotifera and protozoa; the rotifera and protozoa consume algæ and bacteria; the algæ finally nourish themselves by the absorption of soluble substances and gases provided in part by the decomposition of animal and vegetable matter brought about by bacteria.

Oysters feed largely upon diatoms, and microscopic methods have proved useful in the study of oyster culture in the Great South Bay, Long Island, and elsewhere.

MICROSCOPIC ORGANISMS IN WATER FROM DIFFERENT SOURCES

The observations of sanitarians and planktologists show that the microscopic organisms are very widely distributed in nature. They are found in all parts of the world, and under great varieties of climatic conditions. They range from the tropics to the polar ice cap. It has been pointed out above that they appeared on the earth in an early geological age, as evidenced by the traces of their existence found in the various geological strata.

In studying the distribution of microscopic organisms in nature it is convenient to consider the following classes of water separately:

1. RAIN WATER.

2. GROUND WATER.

Springs, Wells, Infiltration Galleries, Infiltration Basins.

3. SURFACE WATER.

Streams, Canals, Ponds, Small Natural Lakes, Artificial Reservoirs, Great Lakes.

4. FILTERED WATER.

5. ICE.

Rain Water. — Rain water is perhaps the purest water found in nature, yet it sometimes contains microorganisms. For the most part they are so minute that an examination by common methods fails to reveal them, but larger forms are sometimes observed.

The study of the organisms found in rain water is really the study of the organisms found in the air. It is worthy of more attention than has been given to it. The presence of organisms, or their spores, in the air may be demonstrated by sterilizing water rich in nitrogenous matter and exposing it to air and light. After a week or two it will contain numerous forms of microscopic organisms which must have settled into the liquid from the air or developed from spores floating in the air.

Rain water collected in a sterilized jar and allowed to stand protected from the air often develops a considerable growth of algæ, usually some *Protococcus* form, showing that the rain has not only taken up the

organisms or their spores, but has absorbed sufficient food material for their growth. Samples of rain water sometimes contain a surprisingly large amount of nitrogenous matter, especially when collected in the vicinity of a large city and at the beginning of a storm.

It has been noticed that vigorous growths of algæ frequently appear in ponds or reservoirs immediately after a rain storm, the growth occurring suddenly and simultaneously throughout the whole body of water. It is suggested that such growth may be caused by the dried spores of algæ that are lifted from the shores of the ponds and scattered through the air by the wind to be washed into the water by the rain. This supposition is in accord with the theory that in the case of certain algæ germination of the spores occurs only after desiccation.

Ground Water. — Ground water is water that has filtered or percolated through the soil. It comes to the surface in springs or is collected in wells or infiltration galleries.

Ground water taken directly from the soil before it has stood in pipes or before it has been exposed to light is almost invariably free from microscopic organisms. Its passage through the soil filters them out. It usually contains an abundant supply of plant food, extracted from the organic and mineral matter of the soil and modified by bacterial action. When the water reaches the light this food material is seized by the microorganisms that are introduced as soon as the water is exposed to the air. One will recall the luxuriant aquatic vegetation at the mouth of some spring or in some watering-trough supplied with spring water. Organisms are occasionally found in ground water supplies before exposure, but their presence usually indicates the admixture of some surface water. With the exception of the schizomycetes (chiefly iron bacteria), the number of organisms found depends upon the exposure of the water to light and air; that is, only as ground water becomes surface water do microscopic organisms develop in it.

Table 1, compiled from the records of the Massachusetts State Board of Health, gives an idea of the organisms found in ground water supplies. Except in the case of spring waters, the figures represent the average of monthly observations extending over one or more years.

Spring waters usually contain no microscopic organisms. Several exceptions are noted in the table — one at Westport, where 455 *Himantidia* (a diatom) were present, and one at Millis, where the water contained 180 *Chlamydomonas* (a green alga) per cc. That these growths were accidental is shown by the fact that in 1893 five examinations of the Aqua Rex Spring at Millis showed an entire absence of organisms.

Well waters also are ordinarily free from organisms, but in some cases *Crenothrix*, the most common iron bacterium, grows abundantly in the

tubes of driven wells. This is particularly true if the water is rich in iron and organic matter while deficient in oxygen. Wells driven in swamps are often affected. The tubular wells at Provincetown are an example. *Crenothrix* is sometimes found there in numbers as high as 20,000 per cc. The water contains more than 0.125 part per million of albuminoid ammonia, and the iron varies from 1.0 to 5.0 parts per million. Many similar cases might be cited. Other fission fungi, such as *Didymohelix*, *Clonothrix*, *Leptothrix*, and *Sphærotilus dichotomus* have also been observed in well waters rich in iron and manganese. *Crenothrix* grows in tufts or in felt-like layers on the inner walls of the tubes. By the deposition of iron oxide on its gelatinous sheath it quickly clogs the tubes and strainers and even the sand around the well points.

Infiltration galleries are, practically speaking, long horizontal wells commonly constructed near some stream or pond. They are similar to wells in regard to the presence of microorganisms. Few organisms other than *Crenothrix* are found, except when surface water gains entrance.

Infiltration basins are infiltration galleries open to the light. The water in them is sometimes affected with algal growths. The infiltration basin at Taunton, Mass., for example, has given trouble from this cause. In October, 1894, there were present more than 1000 *Asterionella* (a diatom) per cc., and they were followed by a vigorous growth of *Dinobryon* (a protozoon). Infiltration basins resemble in many ways open reservoirs for the storage of ground water and are subject to the same troubles from plankton growths.

Surface Water. — The term "surface water" includes all collections of water upon the surface of the earth, such as lakes, reservoirs, ponds, rivers, pools, and ditches.

Table 2 shows that surface waters contain many more microscopic organisms than ground waters, and that standing water contains more organisms than running water.

River Water. — Rivers that do not drain lakes or reservoirs seldom contain large numbers of microscopic organisms, and water supplies drawn from rivers and subjected to limited storage are not often troubled with animal or vegetable growths. This may be true even where the banks of the stream are covered with aquatic vegetation. The organisms found in streams often include a great variety of genera, many of which are likely to be littoral or benthal forms. Their food supply is brought to them by the water that flows by. In quiet slow-moving waters there are free-swimming forms that go in search of their food. Some of these are free-swimming at will or during a part of their life history, and some free-swimming organisms are always found associated

TABLE 1
MICROSCOPIC ORGANISMS IN GROUND WATERS
Standard Units per cc.

No.	Locality	Time	Diatomaceae	Chlorophyceae	Cyanophyceae	Fungi*	Rhizopoda†	Infusoria†	Rotifera	Total Organisms	Zoöglaea
I	SPRING WATERS										
II	Spring in Westport.....	Apr. 21, 1894	455	0	3	0	0	0	1	459	0
III	Aqua Rex Spring, Mills.....	Aug. 27, 1894	1	180	0	0	0	0	0	181	0
IV	Craig Spring, West Springfield.....	May 16, 1893	21	0	0	0	0	0	0	21	16
V	Spring in Ipswich.....	July 27, 1892	12	0	0	0	0	0	0	12	0
VI	Spring in Pepperell.....	Nov. 26, 1894	1	1	0	2	0	0	0	4	0
VII	Massasoit Spring, West Springfield.....	May 16, 1893	2	0	0	0	0	0	0	2	0
VIII	Spring in Ware.....	July 17, 1893	0	0	0	0	0	1	0	1	0
IX	Spring in Medfield.....	Aug. 31, 1894	0	0	0	0	0	0	0	0	0
X	Spring in Pittsfield.....	Aug. 27, 1894	0	0	0	0	0	0	0	0	0
X	Cold Spring, Plymouth.....	July-Dec. 1894	0	0	0	0	0	0	0	0	0
I	WELL WATERS										
II	Tubular Well, Provincetown.....	1894	0	0	0	3130	0	0	0	3130	50
III	Tubular Wells, Revere.....	1894	1	0	0	281	0	0	0	282	—
IV	Large Culling, Well, Marblehead.....	1894	0	0	0	173	0	0	0	173	8
V	Tubular Wells, Hyde Park.....	1893-4	2	0	0	68	0	0	0	70	18
VI	Tubular Wells, Malden.....	1891-3	5	0	1	1	pr.	1	0	8	7
VII	Tubular Wells, Lowell.....	1893	0	2	0	0	0	0	0	2	—
VIII	Tubular Wells, Melrose.....	1894	0	0	0	1	0	0	0	1	—
IX	Tubular Wells, Bradford.....	1893	0	0	0	1	0	0	0	1	—
X	Well at Needham.....	1894	0	0	0	0	0	pr.	0	0	547
X	Well at Fitzwilliam, N. H.....	1893	0	0	0	0	0	0	0	0	0
I	FILTER GALERIES										
II	(Infiltration Galleries)										
III	Filter Gallery at Reading.....	1891-4	3	0	0	3506	0	2	0	3511	726
IV	Filter Gallery at Wayland.....	1891	15	4	1	1766	0	3	0	1779	71
V	Filter Gallery at Whitman.....	1891	pr.†	0	0	137	0	0	0	138	41
VI	Filter Gallery at Framingham.....	1892	1	0	0	217	0	0	0	217	72
VII	Filter Gallery at Braintree.....	1891	0	0	0	137	0	0	0	138	41
VIII	Filter Gallery at Woburn.....	1894	0	0	0	34	0	2	0	36	94
IX	Filter Gallery at Taunton.....	1891	2	0	0	0	0	0	0	2	1
X	Filter Basin at Newton.....	1891-4	86	2	4	0	0	48	1	163	14
X	Filter Basin at Newton.....	1892-4	2	1	0	24	0	pr.	0	16	13
X	Filter Basin at Waltham.....	1892	17	0	0	15	0	pr.	0	17	4

* Including the Schizomycetes. The organisms were chiefly Crenothrix.

† Protozoa.

‡ Present

TABLE 2
MICROSCOPIC ORGANISMS IN SURFACE WATERS
Standard Units per cc.

No.	Locality	Time	Diatomaceae	Chloro- phyceae	Cyano- phyceae	Fungi*	Rhizo- podaf	Infu- soriat	Ro- tilera	Total Organisms	Zoöglea
RIVERS											
I	Stony Brook, Inflow to Basin 3.	1891-2	77	23	43	38	1	9	0	191	97
II	Mill River at Taunton.	July-Sept. 1893	3	25	1	165	1	4	pr.	199	676
III	Merrimac River at Lawrence	1891-4	66	21	2	13	pr.	4	pr.	106	186
IV	Ipswich River.	1892	12	1	0	87	0	74	pr.	106	31
V	Blackstone River at Uxbridge.	1892	17	6	0	3	0	0	pr.	98	364
VI	Sudbury River, Inflow to Basin 2.	1891-2	45	16	2	32	0	3	pr.	128	128
VII	Cold Spring Brook, Inflow to Basin 4	1891	51	pr.†	0	12	0	1	0	77	39
VIII	Nashua River, North Branch.	1893	13	4	2	42	0	6	0	67	810
IX	Taunton River.	1891-3	17	1	2	13	0	2	0	35	58
X	Lynde Brook, Worcester.	1891	17	4	3	2	0	1	0	27	68
NATURAL PONDS											
I	Mythic Lake.	1891-4	1917	199	pr.	18	pr.	172	pr.	2306	128
II	Jamaica Pond.	Jan.-Aug. 1891	1110	103	137	1	1	12	1	1365	174
III	Horn Pond, Woburn.	1891-4	911	302	218	1	1	167	2	1602	65
IV	Fresh Pond, Cambridge.	1891-4	967	95	83	9	1	4	pr.	1159	127
V	Wenham Lake, Salem.	1891-4	897	38	32	0	pr.	32	pr.	999	53
VI	Buckmaster Pond, Norwood.	1891-4	184	83	9	2	1	685	pr.	944	30
VII	Lake Cochichewick.	1891-4	570	33	58	6	2	15	pr.	693	66
VIII	Spect Pond, Malden.	1891-4	171	85	19	1	1	19	pr.	296	93
IX	Lake Williams, Marlboro.	1891	170	17	66	1	0	11	0	268	67
X	Gate Pond, Hudson.	1891-4	110	37	27	1	1	66	pr.	242	38
ARTIFICIAL RESERVOIRS											
I	Haynes Reservoir, Leominster.	1891	3193	0	0	1	0	19	1	3214	155
II	Walden Pond, Lynn.	1891-4	254	238	604	8	pr.	337	1	1302	64
III	North Reservoir, Winchester.	1891-4	1337	35	72	1	1	149	pr.	1396	71
IV	Ludlow Reservoir, Springfield.	1891-4	501	260	96	5	1	96	2	964	103
V	Scott Reservoir, Fitchburg.	1892	691	146	10	2	4	92	3	947	42
VI	Holden Reservoir, Worcester.	1891-4	646	24	6	1	pr.	29	1	707	76
VII	Basin 3, Boston.	1891-4	270	55	23	1	1	12	pr.	362	122
VIII	Basin 2, Boston.	1891-4	99	32	47	5	pr.	4	pr.	187	125
IX	Basin 4, Boston.	1891-4	80	31	3	1	0	5	0	120	43
X	Basin 6, Boston.	1894	55	5	0	0	1	31	2	94	20

* Including the Schizomycetes.

† Protozoa.

‡ Present.

with attached or sedentary forms. In most rivers there are quiet pools or backwaters where free-swimming forms may develop along with the euplankton and benthos. In many streams there are dams that back up the water so as to form large reservoirs. Here luxuriant growths may occur. Thus we find that the water of the many-dammed Ohio River often contains at Louisville and elsewhere such high numbers of diatoms as to have a marked influence on the filters through which the city water is passed.

In a sample of river water, then, one is likely to find littoral or benthal forms that have become detached, plankton and nekton that have developed in the quiet places or in tributary ponds, and spores or intermediate forms in the life history of sedentary organisms. In streams draining large ponds or lakes the water naturally partakes of the character of the pond or lake in which it has its source, and organisms may then be abundant.

The number of microscopic organisms found in rivers is subject to great fluctuations. If the water is rich in food or contains great expanses of backwater, growths often develop quickly and abundantly. Heavy rains that increase the current velocities and cause the backwater areas to overflow may then suddenly wash large volumes of organisms into the stream. Under these conditions the number of organisms collected in a sample may be above the normal. At other times rains may diminish the number of organisms in a sample by dispersion of the plankton and dilution of the food materials. Swift mountain brooks seldom yield appreciable plankton catches. Their turbulence breaks up the fragile organisms and, except for small swampy areas, their backwaters are commonly small in extent.

Table 2 shows that the diatoms are the organisms found most constantly in rivers, but their numbers are small compared with those found in standing water. Some of the chlorophyceæ are often observed. The cyanophyceæ seldom occur. Stony Brook, in Table 2, represents a stream affected by tributary ponds where cyanophyceæ abound. Crenothrix is quite often reported in river waters, but Anthophysa (a protozoön) is often mistaken for it, and this may account in part for the high figures in the table. Animal forms are not common in rivers unless the water is polluted, but when this is the case there may be a downstream succession of protozoa, algæ, rotifers, and crustacea.

Canal Water. — In the slowly moving water of canals and ditches, organisms may develop in large numbers, but conditions are not often such as to cause trouble in public water supplies. The following instance, however, of a veritable water calamity due to growths of organisms in a canal is worth reporting:

On Sunday, July 12, 1896, some of the residents living in the western part of the city of Lynn, Mass., observed that the water drawn from the service taps had a green color. A glass of it showed a heavy green sediment after the water had stood only a few minutes. On the following day the water became worse, and when it was used in the laundry it left green discolorations like grass stains on the clothes. Investigation showed that the stains were caused by *Raphidomonas*, a chlorophyll-bearing protozoön that was found abundantly in the city water. Examination of the four storage reservoirs showed that the organisms were not present there in sufficient numbers to account for the trouble. The water from one of the supply reservoirs, Walden Pond, reached the pumping station by means of an open canal, tunnel, and pipe line. It was in this open canal that *Raphidomonas* was found. The sides of the canal were thickly covered with filamentous algæ, chiefly *Cladophora*. The water in the canal had a dark green color. When a bottle of it was held to the light it was almost opaque and was seen to be densely crowded with moving green organisms. As many as 2000 per cc. were present. Evidently the *Raphidomonas* had developed among the algæ in the canal and had gradually passed into the water from Walden Pond as it flowed through the canal on its way to the city. The trouble was remedied by emptying the canal through its wasteways and cleaning the slopes to prevent further growths. This is the only case on record where *Raphidomonas* has caused trouble, though the organism is often found in small numbers in surface water supplies.

Lakes and Reservoirs. — All quiescent surface waters are liable to contain microscopic organisms in considerable numbers. The standing surface water that is entirely free from them is very rare. It is seldom possible to collect a sample of stagnant water at any season of the year without obtaining one or more forms of microscopic life. They are present not only in the mud puddles in the streets, but in large reservoirs; not only in rain barrels, but in the Great Lakes and in the ocean. The extent and character of the growths vary greatly in different ponds and at different seasons.

As most of our water supplies in America either are derived from ponds, lakes, and impounding reservoirs or are stored in reservoirs when derived from other sources, and as it is in ponds, lakes and reservoirs that microscopic organisms cause the most trouble, it is these bodies of water that will chiefly interest us. As shown in Table 2, all classes of organisms, except perhaps the schizomycetes, are much more abundant in natural ponds and in reservoirs than in rivers.

Filtered Water. — Water that has been filtered, either by slow sand filtration or by mechanical filtration, seldom contains many microscopic

organisms. Their presence in a filter effluent generally indicates that filtration is imperfect. In mechanical filters microscopic organisms are somewhat more likely to appear in the effluent than in slow sand filters. This is due in part to the use of coarser sand and higher rates of filtration and in part to the fact that the organisms become attached to the sand grains near the surface and are carried to the bottom of the filter during the process of washing. They subsequently become dislodged and appear in the effluent. The presence of a few microscopic organisms in the effluent of a mechanical filter, therefore, does not necessarily indicate poor filtration.

Occasionally growths of *Crenothrix* and allied species occur in the underdrains of sand filters. They usually appear where the conditions are such that the water is deprived of part of its oxygen, or where, through leakage, ground water, containing iron and carbonic acid and devoid of oxygen, becomes mixed with the filtered water.

Growths of microscopic organisms often occur in filtered water that is exposed to sunlight in open reservoirs.

Ice. — Algae sometimes become frozen in the ice of ponds. They give the ice a dirty appearance and on decay may cause foul odors months after the ice has been harvested. In artificial ice algae may be concentrated in the "core," so as to produce a noticeable discoloration and taste if the core is not withdrawn.

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CHAPTER II

MICROSCOPIC ORGANISMS AND SANITARY WATER ANALYSIS

The microscopical examination of water is one of that large group of related tests, involving physical, chemical, and biological methods of observation, which as a whole, or in part, constitutes a sanitary water analysis.

The microscope was long used for random studies of the organisms found in natural waters. These studies gave little information upon sanitary problems until the development of chemistry and the biological sciences brought new light to their interpretation and gave a better understanding of their significance. To-day, microscopical examinations constitute one of the most important sources of information upon the sanitary quality of water.

The nature of the evidence deduced is both direct and indirect. Enumeration and identification of the microscopic organisms give direct information upon the following points:

1. They indicate the mass or bulk of living material in the water, with the exception of the bacteria.
2. They establish the character of the organisms making up this material.
3. They denote the immediate effects produced upon the quality of the water by the organisms.
4. They oftentimes throw light upon the history of the water by divulging the presence of adventitious organisms.

Enumeration and identification operate indirectly by aiding in the interpretation of other elements of the analysis. There is a mutual relationship between the microscopical examination and various other tests that logically brings them together in a sanitary water analysis. Not only is the quality of water affected by microscopic growths; it is also true that quality exercises profound influences upon the growths. These effects are noted and measured on the one hand by a knowledge of what the microscope reveals, on the other by the results of physical, chemical, and bacteriological findings.

NATURE OF SANITARY WATER ANALYSIS

In general, a sanitary analysis is designed to give information upon the wholesomeness of water, its general fitness for domestic uses, and the progress of those changes that degrade or improve its quality. To

this end the laboratory analysis comprehends the results of many separate determinations that have to do with physical, chemical, bacteriological, and microscopical qualities. Many of the constituents determined do not measure qualities that are easily defined; they are without special significance when considered alone. Herein lies the complexity of water analysis, which is unlike the analysis of many common substances. Interpretation must be based upon a consideration of all the data obtained. Individual results are often without use except to record fluctuations, and they may have their limiting values modified by other elements of the analysis. Some of the relations of various constituents that are determined by analysis will be pointed out in this chapter, particularly those bearing upon the microscopical content of water.

Necessity for a Variety of Tests. — The scope of the tests employed in a sanitary analysis is purposely broad, for several reasons. In the first place, it is sometimes necessary to base the interpretation of results almost wholly upon data secured from the laboratory, i.e. without the supplementary aids to analysis that will be discussed later. If the fitness of the water for general use is in question such a limitation seriously handicaps the formation of final judgment. Not only must the question of present acceptability from the physical, chemical, and biological standpoints be given consideration; future contingencies must be weighed and a knowledge of the origin of certain constituents must be obtained. The situation can be met best by securing in the laboratory all possible information upon the composition and properties of the sample.

A second reason for using a variety of tests is that the results of one test may modify or amplify the significance of another. The results of all determinations must be considered as a whole, as well as individually. They are not likely to be too comprehensive. One set of tests serves, also, to check the accuracy of others.

Finally, a complete analysis corroborates and extends the knowledge gained from supplementary sources.

Classification of Tests. — The long list of tests common to the subject of sanitary water analysis may be grouped in a variety of ways. Perhaps the oldest and most used system is that resting on the procedures that are required.

Classification According to Procedure. — There are four groups of tests defined by the nature of the procedure employed, as follows:

1. Physical Tests.
2. Chemical Tests.
3. Bacteriological Tests.
4. Microscopical Tests.

Such a grouping brings together determinations having to do with constituents that many times are of widely different origin and that are unrelated in composition or general significance. This is especially true of the first two groups. Turbidity is placed in the first group, but the substances causing turbidity bear little relation, through their origin, composition, or behavior, to those causing color or to those causing taste. Hardness, in the second group, is totally unrelated to oxygen consumed, in the same group; neither does the test for iron give greater or less significance to that for ammonia nitrogen.

The bulk of the tests are thrown into the second group, a heterogeneous one in its relationships. The third and fourth groups contain only a few tests and these are in more logical association. The system as a whole was built up around laboratory procedures without much regard to the ultimate use of the individual determinations.

Direct and Indirect Tests. — Some of the tests have a direct bearing on the quality of the water. Their results quantitatively express the amount of different substances and the intensity of different properties with which the user of the water may be immediately concerned. The *direct* tests are like those for sulphur in coal, magnesium in cement, and tensile strength in steel; they answer directly questions that the user may desire to have answered. Such tests are those for temperature, turbidity, color, odor, taste, chlorides (if high), hardness, iron, iodine, and microscopic organisms. They tell if the water is cool, if it is attractive to the senses, if it is salty, if it is hard, if it is deficient in iodine, and if it contains organisms that depreciate physical quality.

Certain other tests may be called *indirect*, or inferential tests. "Oxygen consumed" and "albuminoid nitrogen" are examples; they supposedly measure the organic matter present, but they do so imperfectly, inaccurately, and only by inference. Oxygen consumed measures organic matter, not directly, but by finding the oxygen used up under prescribed conditions by the organic matter. The albuminoid nitrogen test determines only a portion of the organic nitrogen present, namely that part which is capable of oxidation under the conditions of the experiment. Neither test gives a true idea of the character of the organic matter.

The "bacterial count" is another indirect test. It does not tell the total number of bacteria present in the sample — no known method will do that — but merely the number of bacteria that will grow in a certain time, at a certain temperature, and on a certain medium, at a rate that will produce visible colonies. The test for *Bacterium coli* is only an approximately quantitative determination; it gives an inadequate measure of the number of *Bact. coli*.

Some tests are direct and exact in the measurement of substances

with which they are concerned, but are better classed as indirect because of their inferential use. Organic nitrogen, ammonia nitrogen, nitrite and nitrate nitrogen are all quantitatively indicated by the proper methods, but their mere presence imparts no significant properties to the water. The story they tell of oxidizing and stabilizing changes that have occurred in the organic matter is, however, very significant.

The indirect tests owe their usefulness to their interpretative value. They are used to judge past, present, and future conditions in the absence of suitable direct tests, or when direct tests need substantiating evidence.

Classification According to Substances Revealed. — The most rational system of grouping tests is in accordance with the chemical or biological nature of the constituents that they register. Table 3 shows such an arrangement.

TABLE 3

CLASSIFICATION OF TESTS USED IN SANITARY ANALYSES IN ACCORDANCE WITH
CHEMICAL AND BIOLOGICAL CONSIDERATIONS

Gases and Volatile Constituents	Mineral Matter	Organic Matter	Mineral and Organic Matter	Living Organisms	
				Bacteria	Microscopic Organisms
Odor 1. Cold 2. Hot Taste Carbon Dioxide Dissolved Oxygen Hydrogen Sulphide	Iron Fixed Solids 1. Total 2. Suspended 3. Dissolved Ammonia N Nitrite N Nitrate N Chlorides Free Chlorine Iodides Hardness Alkalinity Incrustants Magnesium Heavy Metals	Color Loss on Ignition 1. Total 2. Suspended 3. Dissolved Total Nitrogen Albuminoid N Oxygen Demand Oxygen Consumed	Turbidity Solids 1. Total 2. Suspended 3. Dissolved H-ion Concentration	Bacteria 1. At 20° 2. At 37° Bact. coli 1. Presumptive 2. Confirmed	Plankton Forms Attached Forms

The person who uses the results of a sanitary analysis to study the quality of a water supply at once finds himself interested in knowing:

1. What is the gross nature of the constituents of the water and their amount?

a. Is there high mineralization?

b. Is there much organic matter?

c. Is there evidence of bacterial contamination?

- d. Are microscopic organisms flourishing in the water?
- e. Are the gases and volatile constituents normal for the type of water?
2. What is the nature and condition of the specific substances making up the different forms of matter present?
 - a. Are they normal to the water?
 - b. Are they stable compounds or do they represent changes that are in progress?

The queries that arise under (1) are answered by considering as a group the results of tests listed under each column heading in Table 3. Those under (2) are answered by scanning the individual results of each group or column of tests.

This preliminary survey of the analysis having been made, further enlightenment comes with a knowledge of water analysis and ability to bring together certain related tests of the different groups. Interpretation of the relations involved affords an extended view of the past history and present quality of the water, from which must be judged the suitability of the source for given purposes.

When tests are thus brought together in logical fashion greater information is presented to one possessing only a moderate knowledge of the subject; also, interpretation of results is much more easily understood by the lay mind if it has as its basis such an arrangement.

Choice of Tests. — A sanitary analysis may or may not call for the use of all the tests given in Table 3. A complete study of sanitary quality would call for nearly all the tests; oftentimes analyses are made to obtain information on particular phases of quality. In such cases a choice of tests must be made that will provide the required data without useless expenditure of time and effort. Water analysis is not a uniform procedure. Sometimes a single simple test may be all that is required; at other times the analysis must be intensive and complicated. To tell what tests are *necessary* and *sufficient* requires good judgment.

Direct and Interpreted Information. — As far as possible, a choice should be made that will answer directly the purposes of the examination. Some questions cannot be answered by direct tests; recourse must then be had to those that have interpretative or inferential value, the indirect tests. The latter demand more knowledge and skill in their application than do the former.

If it is desired to know if the water is corrosive and if it contains lead, determinations of carbon dioxide, hydrogen ion concentration and alkalinity will give direct information upon the solvent properties, and that of lead upon the presence of this metal and its amount.

If the effect of storage upon odor and appearance is sought, determina-

tions of taste and odor will give direct evidence upon this point, and a microscopical examination will tell if organisms are present that are causing or may cause odor. Tests for color and turbidity will directly point to changes in appearance.

Should information be desired upon the nature and amount of organic matter present in a water sample, tests for loss on ignition and color will measure the amount and tell if extracted vegetation contributes largely to it. Microscopical examination will show whether microscopic organisms make up part of the organic matter. Indirect tests will be the only ones to give an idea of the composition and stability of the organic content. That for albuminoid nitrogen will measure the easily decomposed nitrogenous matter; that for oxygen consumed will principally indicate carbonaceous organic matter; the oxygen demand will give the total amount of oxygen required for the biochemical oxidation of the organic matter. By inference, judgment may be formed as to the stability of the organic compounds and the relative proportion of nitrogenous matter that is present. Bacteriological and microscopical examination will give evidence of interpretative value in judging whether or not sewage has contributed to the organic content.

Tests for Wholesomeness. — Wholesomeness is one of the most important qualities of water to be questioned. As far as mineral substances are concerned a fairly direct answer may be obtained from a sanitary analysis. There are, however, no direct tests for pathogenic organisms in water that can be conducted with assurance of accurate results. Reliance must be put upon data that measure the opportunity that the water has had to become infected by pathogenic bacteria, and the possible changes that may have occurred in the bacterial content before the sample was taken. To this end a bacteriological and microscopical examination must be made and, also, tests that deal with the organic and mineral contents and that present a history of the changes subsequent to any contamination. The organic and especially the bacteriological tests measure sewage contamination more definitely than the mineral tests, but the results must be confined more closely to the given time and place. The mineral tests, although they are less definite, represent a greater integration of conditions, both in time and volume of water. The two groups thus supplement each other. All the tests acquire added import from a knowledge of field conditions.

Tests to Measure Purification. — The measurement of the changes that are effected by artificial processes of purification calls principally for the use of direct tests and for the bacteriological and microscopical examinations. Determinations of color, turbidity, odor, hardness, alkalinity, iron, dissolved gases and microscopic organisms, all give

direct answers to questions regarding the amount of certain substances that have a significant influence upon quality. In the case of water rich in organic matter, the nitrogen determinations and that of oxygen consumed may be useful.

Studies involving self-purification processes make greater use of tests that measure changes indirectly or by inference. Such studies are largely concerned with the disposition of organic matter and living organisms, for which there are few direct tests. The microscopical examination is one of the direct means of measurement that has surpassing importance in connection with the pollution and self-purification of water.

Other Factors Dictating Choice of Tests. — It is not always possible to select in advance a list of determinations that will give the required information for a sample of unknown quality. It is usually the case that some of the tests will have little value; others will yield data that need to be amplified by tests not already chosen. The best practice under such circumstances is to make a few tests that will give direct evidence on matters of importance; then to add any others that this approximation of quality may suggest as being necessary. The proper combination of tests is a matter deserving careful attention, for there are numerous supplementary relationships among tests that give great aid to the analyst.

It should always be borne in mind that the elapsed time between collection and analysis of a sample indicates whether or not determinations of perishable constituents should have precedence over others.

An analysis should be so conducted as to give internal checks, whenever possible, on the results of determinations. These check tests give assurance of consistency and accuracy. For example, odors may in some instances be verified by microscopical examination; supersaturated values for dissolved oxygen and negative results for carbon dioxide may also be confirmed by the same procedure; the fixed solids should be greater than the sum of hardness, chlorides, and other mineral constituents; total organic nitrogen should exceed albuminoid nitrogen. Obviously, these check tests are unnecessary when the character of the sample is well known from repeated analyses.

RELATIONS OF MICROSCOPIC ORGANISMS TO THE ANALYSIS

The relations that microscopic organisms bear to other constituents determined by a sanitary analysis are numerous. The microscopist, the chemist, and the bacteriologist here have mutual interests. They should have a clear understanding of the interlocking significance of

different elements of the analysis; without this knowledge they lack the breadth of view that is necessary for the solution of many problems connected with sanitary studies of water.

Interpretation of Typical Analyses. — The following pages will deal briefly with some of the dual relations existing between the test for microscopic organisms and some of the other common tests of a water analysis. Additional information on the subject will be found in Chapters III, VIII, XI, and XII. The discussion will take the form of an interpretation of a series of hypothetical sanitary analyses made upon water samples collected from different stations on two catchment areas. Consideration will be given principally to those determinations which have to do with constituents that have influenced the growth of microscopic organisms and with those which have been subject to change as a result of the increase in organisms. The results of the analyses appear in Table 4; a sketch map of the stream courses is presented in Fig. 1. The location of sampling stations is given on the map.

It should be noted that the stream from the northwest has two branches: the one to the west rising in wooded upland country devoid of habitations and then flowing through an agricultural region; the one to the east rising in a swamp contiguous to sparse population. The two branches unite to enter an impounding reservoir. This stream receives drainage only from rural communities. The stream from the northeast rises also in upland country, but later cuts through the center of a city, the principal sewer outfall of which is just below the city limits and discharges into the stream. Samples from each station on the two streams will be considered individually and in their relations to samples from upstream stations.

Upland Stream (Station A). — The clean water at Station A contained only 50 standard units of microscopic organisms per cc.; these were chiefly species of diatomacæ having widespread distribution in such water. The numbers were too few to influence the quality of the water. Carbon dioxide, dissolved oxygen, and the nitrogen values were normal for waters originating in wooded upland regions. The very faint vegetable odor was due to a small amount of organic matter of plant origin.

No substances were present in large enough amounts to stimulate the growth of either plant or animal forms of life; furthermore, the recent origin of the stream and its rapid movement did not offer opportunity for the breeding of microscopic organisms.

Stream below Farm Lands (Station B). — Somewhat different conditions were found at Station B, downstream from Station A. The stream was larger, lower in velocity, and added time had been allowed

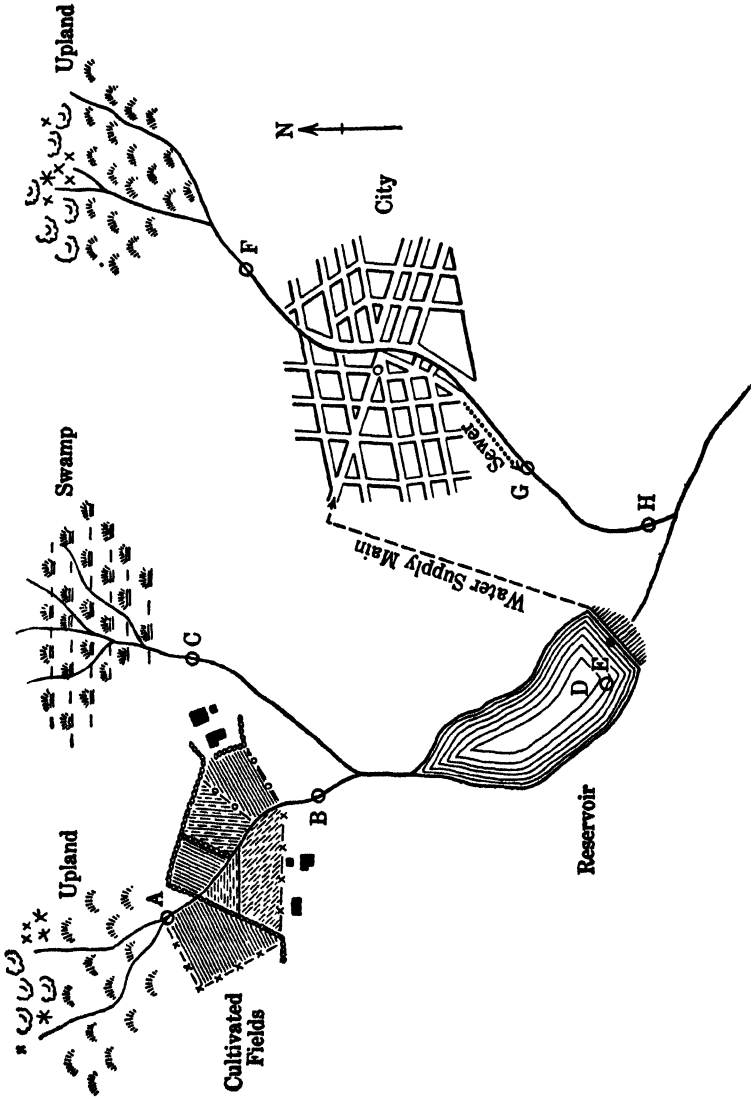


Fig. 1. — Map Showing Location of Sampling Stations. See Table 4.

TABLE 4

TYPICAL ANALYSES OF SURFACE WATERS

FIELD RECORD										Report of TYPICAL ANALYSES OF SURFACE WATERS									
										To.....(Accompanied by sketch map.).....									
										Date.....Sept. 2, 1928.									
Sample Number		Time of Collection		DESCRIPTION OF SAMPLE				GASES		ODOR									
		Date 1928	Hour					Temperature	Dissolved Oxygen			Carbon Dioxide	Cold	Hot					
A		Sept. 1	1 P.M.	Upland Stream near headwaters Stream below cultivated land and farm houses Stream below city (surface sample) Lower end of reservoir (bottom sample) Lower end of reservoir (surface sample, 60') Upland stream above city Upland stream below outfall of city sewer Sewage at outfall of city sewer; in fresh condition				Parts per Million	Per cent of Saturation	1.5	1v	1v							
B		"	2 "					9.48	100	2.0	1v	1v							
C		"	3 "					6.8	88	5.0	3v+2M	4M							
D		"	4 "					7.0	115	0	2v+1.3g	4g							
E		"	4 "					10.12	0	25.0	4g	3d+3S							
F		"	5 "					5.0	100	10.0	1v	1v							
H		"	6 "					9.48	40	2.0	2M+3d	3M+3d							
G		"	5:30	0.85	10	25.0	4d	5d											
ORGANIC MATTER										LIVING ORGANISMS									
Loss on Ignition										BACTERIA									
										MICROSCOPIC ORGANISMS, Std. Units per cc.									
Color	Total		Sus-pended		Total		Oxygen Con-sumed		No. per cc.		Tests for Bact. Coli.		Total Organ-isms		Principal Groups				
	Sus-pended	Dis-solved	Sus-pended	Dis-solved	Total	Total	Total	20°	37°	.01 cc.	0.1 cc.	1.0 cc.	10 cc.	Dia-tom-osee	Cyano-phyceae	Proto-zoa	Fungi		
12	10	1035	2.1	50	10	0	0	40		
18	15	885	3.0	350	150	0	0	175		
125	30	210	16.5	390	50	0	0	300		
55	21	320	11.0	150	20	0	0	300		
200	46	155	18.0	650	70	0	0	3200		
5	7	2020	1.8	35	4	0	0	100		
13	40	750	14.0	9000	7500	0	0	20		
22	380	185	205	5,000	62.0	2.0M	1.8M	+	+	1000+		
										MINERAL AND ORGANIC MATTER									
Tur-bidity		Solids		Fixed Solids		Chlo-rides	Hard-ness	Alka-li-nity	Incrus-tants	Iron	Magnes-ium	H.I.C. pH	Nitrogen as						
		Total	Dis-solved	Total	Sus-pended								Dis-solved	Free Ammonia	Ni-trates	Ni-trogen			
2	30	2.8	12.0	10.0	2.0	0.1	6.7	0.20		
3	42	2.6	16.0	11.0	5.0	0.2	6.8	0.75		
3	52	2.4	10.0	9.0	3.0	0.4	6.6	0.35		
8	48	2.6	13.0	10.0	4.0	0.2	6.8	0.75		
15	79	2.6	13.0	10.0	3.0	0.2	6.6	0.75		
1	49	2.6	22.0	20.0	7.0	0.1	6.9	0.15		
25	110	12.0	42.0	32.0	10.0	0.5	6.7	0.15		
220	750	250	500	23.0	80.0	95.0	10.0	1.5	7.0	0.10		

Crustacea, such as Cyclops and Daphnia, the rotifer Anuræa, and a few protozoa were also noted.

The numerous plankton population affected the quality of the water in several ways. The physical appeal, or attractiveness, suffered depreciation, principally because of odor and taste. The moldy and grassy odors recorded were characteristic of growths of cyanophyceæ. Such odors are more unpleasant than the ones noted in the streams above the reservoir. Quiescent storage tends to reduce turbidity, but in this sample the turbidity increased over that of the upper station waters, the value of 8 parts per million being 5 or 6 parts in excess of what would be expected from the turbidity at Stations B and C. The color of the water was slightly greenish, indicating that part of the color was due to the algæ present.

Photosynthesis was responsible for the dissolved oxygen being present in excess of saturation requirements; the same phenomenon resulted in complete assimilation of free carbon dioxide. Among the mineral constituents the alkalinity was partly converted to normal carbonates, due to removal of carbon dioxide from bicarbonates by the actively growing plant organisms. The latter also made use of the nitrates of the incoming water, reducing the nitrates in the reservoir to the low figure of 0.10 part per million. The hydrogen ion concentration, or pH value, was markedly influenced by the removal of carbon dioxide and formation of normal carbonate. Analysis recorded a pH value of 7.4, indicating a change to alkaline condition from the acid reactions obtained at the upper stations.

The ammonia nitrogen owed its comparatively low value to the fact that the microscopic organisms were in a thriving state and were not undergoing decomposition. The low bacterial counts were also evidence of an actively multiplying plankton population. The reduction in numbers of bacteria from those present in the stream waters above, and the absence of *Bacterium coli*, were partly due to the higher pH value of the water, the latter being commonly known to exert a bactericidal effect.

Albuminoid nitrogen was very high, but should not be taken as an index of the potential fertility of the water inasmuch as the source of the nitrogen was largely in the cells of living microscopic organisms. The value for oxygen consumed also reflects the presence of the organisms; 6 to 8 parts per million would be the amount of oxygen consumed in the absence of microscopic life for an average water of equivalent organic content.

Lower End of Reservoir, Bottom Sample (Station E). — The sample from Station E was taken at the same location in the reservoir as that

from Station D, but at a depth of 60 feet. It illustrates the striking differences between top and bottom water during the period of summer stagnation. Mixing of the upper waters of such a reservoir occurs to a depth of 30 or 40 feet; below this point there is no mixing until the fall overturn. There is an accumulation of organic matter in the bottom waters and anaërobic conditions prevail there.

The microscopic life reflected the absence of light and oxygen, which are prime requisites for development of algæ. Only 400 standard units of microscopic organisms were observed; of these, 100 units were made up of diatomaceæ. The genera were those noted at the surface, but the cells were in an attenuated state, or else dead, consisting almost wholly of siliceous skeletons. They had been carried downward by gravity and did not represent the normal life of the zone of stagnation. Fungi were found in considerable numbers, the genus *Crenothrix*, one of the filamentous higher bacteria, making up 225 standard units of the count. This form is a frequent inhabitant of both stagnant and running water that contains organic matter and iron salts.

Both organic matter and iron were present in large amounts, the former being indicated by the tests for color, loss on ignition, albuminoid nitrogen, and oxygen consumed. Bacteria were present in considerable numbers; their activity was manifest from the ammonia nitrogen content, the carbon dioxide, and the absence of oxygen. The latter, together with the odor of hydrogen sulphide, indicated the anaërobic nature of the environment. Very few microscopic organisms can maintain themselves under such conditions. The acid reaction of this water is worthy of note, a pH value of 6.0 being recorded, largely due to the high concentration of carbon dioxide.

The microscopic organisms were not numerous enough measurably to affect the analysis. Iron-encrusted sheaths of *Crenothrix* doubtless added to the turbidity and color and to some extent increased the organic matter and albuminoid nitrogen. They did not appreciably alter the amount of the mineral constituents except by precipitation of some of the iron.

Upland Stream above City (Station F). — The sample from Station F, on the stream draining the catchment area to the east, showed that the water from this stream was somewhat higher in hardness than the one to the west, that it was physically clean, and that it contained few living organisms, either bacteria or other microscopic forms. The organic and mineral impurities of the water contributed very little to either immediate or potential fertility toward plankton life. Only 25 standard units of organisms were found, of which 20 were diatomaceæ.

Sewage (Station G).— At Station G was the main outfall of the city's sewer system. The sample taken at this point represented a composite of combined sewage from a city with small manufacturing interests and served by a water supply drawn from the impounding reservoir located on the catchment area to the west. The sewage was in fairly fresh condition, containing dissolved oxygen to the extent of 10 per cent of saturation and 0.10 part per million of nitrogen as nitrate.

Inspection of the analysis shows typical results for such sewage; it carries a great store of raw food that becomes available for plant and animal life after various katabolic and anabolic changes have taken place. In addition there are in abundance constituents such as the bacteria, together with carbon dioxide and ammonia compounds, that are available without change to stimulate the growth of certain microscopic organisms. The utilization of these compounds as food rests upon establishment of the proper environment.

Raw sewage flowing in underground conduits allows the development of comparatively few forms of life except the bacteria. The sample from this station contained only 150 standard units of microscopic organisms, of which 25 were the siliceous skeletons of diatomacæ that had their origin in the city water. There were 70 units of protozoa, principally Arcella, and 50 units of mold hyphæ, the latter probably washed from the walls of the sewers.

Stream below Sewer Outfall (Station H).— Below Station G the complexion of the stream changed markedly. Above this point the water was comparatively clean, pollution from the city being of a character and of an amount that did not greatly debase the physical and chemical qualities observed in the water from Station F. With the entrance of a large volume of sewage at Station G, a heavy load of suspended and dissolved impurities, organic and inorganic, was introduced into the stream. The forces of self-purification soon set to work to care for this burden and in due time materially altered downstream the conditions observed at the point of sewage discharge. A sample taken downstream at Station H will serve to show the qualities of the stream water at a point where active decomposition had noticeably slackened and where recovery to clean conditions had begun to manifest itself. The station marked the beginning of the *zone of recovery*.* Dissolved oxygen was present to the amount of 40 per cent of saturation; the odor of the water was musty and disagreeable and nitrates had increased to 0.20 part per million.

The microscopic content of the sample consisted of 200 standard units of cyanophyceæ, principally Oscillatoria; 500 units of protozoa,

* See Chapter XII.

Paramecium, Colpidium and Vorticella; 150 units of schizomycetes, Sphærotilus natans and Beggiatoa; and 150 units of rotifera and crustacea, Brachionus and Rotifer, and Daphnia and Cyclops. Some of these forms were typical for the environment in which they were found; others had probably been carried downstream from points above. All were important agents in carrying forward the processes of digestion and stabilization of the organic impurities that were introduced into the stream with the sewage.

The marked reduction in turbidity, reaëration of the stream, and presence of carbon dioxide, all of which were apparent at this station, made possible the development of Oscillatoria, one of the first indicators of recovery to clean conditions. The number of cyanophyceæ and other algae would be expected to increase beyond this point. The effect of their growth would be noted in an increase in oxygen, a reduction in carbon dioxide, and a decrease in the hydrogen ion concentration.

The relatively large number of protozoa observed owed their presence to the abundance of food available in the polluted stream. They are active bacteria-eaters. Below the sewer outfall the number of bacteria was very large. The changes wrought as a result of their saprophytic existence are indicated in the analysis by total and albuminoid nitrogen and oxygen consumed, which registered a marked reduction in the amount of organic matter in passing from Station G to Station H. At Station H the number of bacteria was conspicuously lower than upstream in the vicinity of the sewer outfall, due to a scantier food supply and to the depredations of the protozoa.

It is probable that the maximum density of protozoön forms prevailed at a point upstream from Station H and nearer to that portion of the stream that contained the densest population of bacteria. At Station H the rotifera and crustacea were in evidence; these are animal forms that require, in general, a more highly oxygenated environment than the protozoa and that include the protozoa as a portion of their food. They also ingest small particles of organic detritus and so aid in the general cleaning-up process that Nature applies to polluted water courses.

The fragments of Beggiatoa and Sphærotilus natans that were reported in the microscopical examination were not indigenous to the zone in which they were recovered. Beggiatoa is a sulphur-loving organism and is usually found where decomposition is active and hydrogen sulphide plentiful, namely, in the *zones of degradation and active decomposition*. Sphærotilus natans, one of the higher bacteria, is a true saprophytic organism and will be found where lower forms of bacteria find abundant pabulum, that is, where proteolysis and fermentation are going on. It will generally be found in tufted masses and not free-

floating in the water. The specimens of these two schizomycetes were carried by the current of the stream to Station H after production and growth in foul upstream water. They serve as indicators of polysaprobic conditions that prevailed between Stations G and H. When taken into consideration along with other organisms found at Station H and along with the quality of the water at that point, as shown by analysis, these members of the class Schizomycetes offer testimony to the magnitude of the purification that had been effected prior to the arrival of the stream water at Station H.

Ground Water. — An analysis of ground water is not included in the typical analyses given above. Ground waters are peculiarly free from nearly all forms of microscopic organisms as drawn from their underground sources. Chlorophyllaceous plant forms are deprived of necessary light, although food is plentiful, for ground waters commonly carry an abundance of carbon dioxide and nitrates. Animal organisms find a great scarcity of proper food in these clean waters.

Among the fungi there are a few organisms, other than the true bacteria, that frequently infest ground waters. These are members of the family Chlamydobacteriaceæ, or higher bacteria, filamentous forms surrounded with a sheath. They are sometimes called the "iron bacteria," because of their association with considerable amounts of iron. *Crenothrix*, *Leptothrix* and *Didymohelix* are typical genera of this family; of these *Crenothrix* is most commonly encountered, sometimes in deep wells, but more often in shallow wells and infiltration galleries. The filaments of the iron bacteria grow attached to well and pipe walls and in the voids of porous strata. They are found in the water after their death or when the velocity of flow has torn them from their points of attachment.

Water from underground sources that are infested with the iron bacteria will contain from a few to a great many standard units of the organisms. They are liable to affect the quality of the water in several ways. When accompanied by ferric oxide or hydroxide, that has been precipitated around the filaments, a visible turbidity of several parts per million may result and the water may have a milky opalescence or a rusty iron color. The odor will vary from faint "musty" to decided "fishy" or "disagreeable," depending upon the organic matter present, the mass of the filaments, and their state of disintegration. Dissolved oxygen will remain practically absent, — the usual condition in ground waters, — and carbon dioxide will be high in amount. The iron bacteria do not utilize carbon dioxide, but may add to it when their filaments decay. Iron will usually be present to the extent of several parts per million; any that occurs in the sheath of iron bacteria

will be in the insoluble ferric condition. Albuminoid nitrogen and oxygen consumed will be increased by either the living or dead cells. Ammonia nitrogen will be increased by bacterial decomposition of the dead organisms, whether they remain attached underground or are detached and appear in the water. Reduction of nitrates by iron salts may also add to the ammonia nitrogen content.

The characteristics of ground waters are often such that they favor the growth of iron bacteria. Dissolved oxygen will be low or absent and carbon dioxide will be high, usually between 10 and 100 parts per million. The color will be low but the total organic matter will be considerable in amount, as indicated by the oxygen consumed, albuminoid nitrogen, and usually the ammonia nitrogen tests. The iron bacteria apparently live a saprophytic existence and require organic food. True or lower bacteria will be few in numbers; consequently they offer little competition for the organic matter. Iron will be abundant in the soluble ferrous condition, although it is not required for food purposes. Other mineral constituents, including the nitrates, will vary widely; they appear to exercise little influence upon the growth of the iron bacteria.

SUPPLEMENTARY AIDS TO ANALYSIS

Any attempt to render an opinion as to the sanitary quality of water, if it is based entirely upon laboratory findings, is exposed to the error of forming judgment without complete knowledge of the situation. Investigation should supplement examination, for there are many factors of environment and of past experience with the water that may throw a flood of light upon its supposed quality as established by analytical data.

There should be inspections in the field, which will yield information as to sources, terrain, pollution, rainfall, etc. In the library and the office, search should be made for previous analyses of the source and for any reports or comments upon it. Statistical investigation should also be undertaken to determine any possible influence of the water upon health. The extent of these supplementary studies will be determined by the exigencies of the situation and by the importance attached to matters of sanitary quality.

Examination in the laboratory definitely establishes the presence of certain substances or organisms, as hardness-forming compounds or bacteria, and records properties such as odor, taste, and oxygen-consuming power. It also yields quantitative figures that make possible precise measurement of the amounts of various substances present; it is the basis of the appraisal of quality. The supplementary aids to

analysis, on the other hand, will yield largely qualitative data, the value of which is interpretative. They will explain sources and fluctuations of the substances registered by laboratory tests and will reflect the results of "clinical" experience in the use of the water.

When all the information has been brought together that bears upon the substances in the water and its qualities, and that relates to its source, history and experience in use, the analyst is fortified with facts that make possible both the broadest interpretation of existing sanitary quality and the prediction of future departure from this quality.

The Field Survey. — There is no better way of estimating what the results of analysis will show than to investigate conditions in the field. The value of such an inspection will, however, vary directly with the qualifications of the person making the survey. The capable observer must have a knowledge of water if he is to judge those matters that vitally affect it. Such knowledge rests upon a familiarity with stream courses, flow of underground water, geological structures, and meteorological conditions, with the substances that pollute water and their possible sources, and with contamination, its significance and the devious routes it may pursue. Above all, he must possess an abundant store of common sense and good judgment that will deter him from false leads and enable him to put a proper estimate upon values.

An analyst was once called upon to investigate the field conditions surrounding a highly colored stream that was used for drinking purposes and had been shown by laboratory tests to be badly contaminated. He was taken by a previous investigator to a barnyard on the headwaters, where there was a large pile of manure, and was told that the leachings from this compost were responsible for the contamination and the color in the water, and that the latter was nothing more than stable extract. Inspection divulged the presence of a small branch of the stream near the barnyard, but the organic material contributed by the latter was a mere drop in the bucket compared with that coming out of swamps along the main stream course. Likewise, the bacterial contamination contributed by run-off from fields fertilized with human and animal waste was a far more serious matter than the contamination that entered from the barnyard. This was, perhaps, an extreme case; nevertheless it affords an example of an investigator led astray by finding one bad situation, the value of which in the whole problem he failed to estimate accurately.

The scope of the field survey will be dictated by circumstances; it may be either broad or limited. For surface waters it will ordinarily include the following:

1. Character of the soil and surface rocks.
2. Character of the catchment area, whether flat or steep; nature of the cover, whether forest, pasture or cultivated land.
3. Population density, sewered population, number and character of industries, railroads, and highways.
4. Disposition of excreta, sewage, and wastes; their proximity to the water intake.
5. Protective measures: flood control; adequacy of storage; control of fishing, boating, and swimming; location of intake; policing; analysis of the water; control or treatment of sewage or wastes.
6. Purification processes: disinfection, filtration, etc.; size of units, control, and efficiency under all conditions.
7. The distribution system: nature and condition of pipes and pumps; cross connections; protection of water as distributed to consumers.

Ground waters usually require investigation of the following items:

1. Nature of the overlying and underlying geologic strata.
2. Size of catchment area and depth to the water table.
3. Nature and distance of sources of pollution: slope of ground-water table; possibility of known sources of pollution being an element of danger.
4. Existing hazards from surface-water pollution: faulty casings or abandoned borings.
5. Protective measures: control or treatment of sewage and wastes; disinfection, filtration, etc.

When a competent person has gathered in the field all the information of the above character that is pertinent to a particular case, facts are at hand that indicate the sanitary aspects of a water supply and make possible an estimate of its hygienic quality. The fullest measure of use for such data comes, however, from their assistance in interpreting the results of laboratory tests, whether the latter have to do with physical, chemical, bacteriological, or microscopical properties. In fact collateral evidence of this kind is in most cases indispensable to forming final judgment of quality. The sanitary survey also brings great assistance to the interpretation of morbidity statistics.

A single survey, like a single analysis, shows conditions at the time of its execution. Changes are likely to occur that greatly modify the original findings. These may be brought about by natural or artificial forces. Population may increase, new sources of contamination may be established, geological structures may fail. Repeated checks upon field conditions are needed to provide new information and to follow developments, the trend of which proceeds slowly but which may have great significance.

The Test of Experience. — A sanitary analysis of water is intended to furnish, among other things, evidence of its wholesomeness or unwholesomeness. In so far as the presence of poisonous compounds or the likelihood of the presence of disease organisms is concerned, this end is usually attainable. There are times, however, when analysis does not provide the expected protection. In some unfortunate instances this state of affairs has come about from placing reliance upon laboratory control and finding it derelict in its functions; or, perhaps, a false sense of security has been founded upon analyses made long in the past. Epidemics of serious diseases, like typhoid fever, have resulted from such misplaced confidence; communities have been afflicted with lead poisoning for the same reason.

Often, when laboratory examination has not provided the proper safeguard, the deficiency has been due to failure of methods to indicate or detect harmful properties. For instance, some of the epidemics of the milder water-borne diseases have not been accompanied by evidence of deteriorating bacterial quality. Similarly, in the past the high incidence of goiter in many communities did not reflect the part played by the water supply, for the reason that analysis was not concerned with the iodine content and did not, therefore, register its deficiency. We now have a better appreciation of the influence of this element upon goiter; wherever the disease prevails to any extent, analysis is likely to furnish corroborative evidence of the cause.

There are doubtless other constituents and properties of water the hygienic significance of which is not yet known. Analysis does not ordinarily deal, by any means, with all the elements of water quality, and medical and sanitary science is constantly adding new knowledge that has to do with previously neglected substances. So it is that the test of experience, — the clinical manifestations from use of a water, — may give valuable supplementary information concerning the effect upon health. This is the final and a direct, although *post facto*, test of wholesomeness.

To apply the measure of such a test it is necessary to collect facts from different sources and to study and apply them with caution. Published morbidity and mortality statistics may provide required data; oftentimes the investigator will find it necessary to compile his own statistics by consulting office records of physicians, or by a canvass of individuals among those using the water. The desired information is likely to be widely scattered and difficult to coördinate.

Frequency of Analysis. — There is no better way of extending the usefulness and significance of analytical results than to increase the frequency with which they are collected. It is a common mistake,

especially among laymen, to suppose that an analysis of water, like that of rock material, represents fixed quality. Nothing could be farther from the truth. The greatest weakness of a water analysis lies in the fact that it shows conditions at the moment of sampling, and does not record past changes or those which may come in the future. For some purposes a single analysis of water suffices; if an accurate estimate of hygienic quality is the end in view, both past and present analyses should be available. Interpretation of sanitary analyses should establish the trend of slow changes and make possible predictions as to future conditions. This cannot be done safely without a knowledge of what has happened in the past.

A well-executed field survey compensates partially, but not wholly, for a paucity of analytical data, in that it presents the possibilities of future happenings. It does not show, as do repeated analyses, the actual fluctuations in quality that occur from day to day or month to month. Frequent samples give mean values by which the departure from normal quality may be measured. They fix, also, the extremes of quality, thus indicating occasional hazards of use and dictating safeguards of practice.

REQUISITES OF RELIABILITY

In order that water analyses may be reliably and effectively used there are certain requisites that must be met, namely:

1. The sample must fairly represent the water from which it is taken.
2. There must be no substantial change in the sample between the time of collection and the time of analysis.
3. The tests made must be adequate to furnish the desired information.
4. The tests must be made accurately by recognized methods.
5. The results must be presented with adequate interpretation and in a manner that will be illuminating to those not thoroughly familiar with the subject.

Errors of Sampling. — There are various ways in which a sample may fail to represent the body of water from which it is taken. These sampling errors fall into three classes.

Errors of Collection. — There may be errors of collection due to the use of unclean, unsterilized or improper kind of collecting apparatus or container; careless handling of the apparatus or container; unclean hands; improper manipulation; insufficient amount of sample; erroneous labelling.

Errors of Inequality. — There may be natural errors due to inequality of the body of water. The waters of lakes and reservoirs show differences between top and bottom, between the inlet and outlet ends, between windward and leeward sides; the water of streams shows differences between top and bottom and between mid-stream and shore; the waters of harbors and tidal estuaries show differences between channels and shallows, and are influenced by inflowing streams, by wind action, and by waves; the water supplied to a city through a reticulation of pipes shows differences at various taps, samples from "dead ends" being sometimes quite different from those collected from taps near the larger mains.

Errors Due to Changes in Course of Time. — There may be natural errors due to changes in the water at the sampling point in the course of time. River waters fluctuate greatly in quality according to the conditions of flow, these changes being largely influenced by rainfall, by melting snow, by ice, and by summer drought. Harbor waters show tidal differences between ebb and flood tides, neap tides and spring tides. Filter effluents may vary from hour to hour; sewage may vary in quality from minute to minute; trades' waste, especially, may vary irregularly in both quality and quantity.

The subject of natural errors that are due to inequalities of bodies of water and to changes at different times and under different conditions is one deserving far more attention than is usually given to it. The general principle should apply that the greater the inequality of the water, the more numerous should be the points of sampling; and the greater and more rapid the time changes, the more frequent should be the collection of samples. Not all the substances revealed by analysis are subject to the same inequality of distribution or to the same change with time. In a reservoir the chlorides or the hardness may remain very constant in value, while the plankton or the bacteria may fluctuate by weeks, days, or hours. It is not necessary, therefore, to consider all tests in the same light.

Errors of Transportation. — Errors incident to transportation may be due to the character of the container, the method of packing, changes in temperature, agitation of the sample, and the time that elapses between collection and analysis. All these changes are influenced by the character of substances in the water. During the interval between collection and analysis, dissolved oxygen may be used up by organic matter, carbon dioxide may escape, colloidal matter may become partly aggregated, bacteria may die or increase in number, the odor may change, delicate plankton organisms may disintegrate. These and other changes that may occur alter the character and amount of substances in the sample.

Errors of transportation may be greatly reduced, if not overcome, by the exercise of due precaution. The allowable lapse of time between collection and analysis varies somewhat with the character of the sample and the constituents that it contains. The limits established by "Standard Methods for the Examination of Water and Sewage" should not be exceeded. Errors of transportation can be avoided by analyses in the field. This is one of the reasons for encouraging the use of field tests.

Adequacy of Tests. — It has previously been said that the making of a sanitary water analysis seldom calls for the use of all the tests named. To conduct unnecessary tests involves a waste of time, effort, and money and makes the report of the analysis less concise and direct. The number of determinations must be adequate to serve the purpose of the examination, the following points being kept in mind, — the use to which the water is to be put, the comparisons that need to be made and the general nature of the whole problem. The selection of tests will be governed by a knowledge of such matters as have already been discussed on page 28.

Standard Methods of Analysis. — One of the prime requisites for a water analysis is that the procedures used shall conform to accepted practice. Many of the determinations are of such a character that they may be carried out by more than one method; the values obtained will depend upon the method chosen, that is, upon the conditions of the experiment. This possibility at one time led to great confusion and indiscriminate results. Every analyst exercised his prerogative of choice and thought his procedures the best.

Order was finally brought out of this chaotic state by acceptance and publication of a manual for water analysis, "Standard Methods of Water and Sewage Analysis." The book was a committee report, sponsored by the American Public Health Association,* and has done more to put water analysis upon a systematic and rational basis than any work before or since published. It appeared in 1905 and is now in its sixth revised edition. The procedures therein laid down have been widely accepted and form the standard practice of the day. Therefore, it behooves the water analyst to follow them unless there is good reason for not doing so. Any departure from these recognized methods should be indicated.

Special emphasis is attached to the importance of reporting analytical results in terms of units commonly employed for the purpose. There are few occasions, for instance, when there is justification for the use of

* "Standard Methods for the Examination of Water and Sewage" is now sponsored, approved, and published jointly by the American Public Health Association and the American Water Works Association.

any form of expression other than "parts per million" (p.p.m.), in reporting the results of chemical tests. In some cases "grains per gallon"* is more convenient, particularly in connection with purification processes. Parts per million is, however, the standard expression that is almost universally used; precedent, personal whim or any trivial reason should not be allowed to set up the use of an expression that is obsolete or uncommon. The units used in reporting microscopical findings are discussed in Chapter VI.

Presentation of Results. — The final requisite of a sanitary analysis, one that concerns its reliability and largely controls its usefulness, is that the results of the various tests be so arranged in the report as to display the constituents of the water in the most orderly and most intelligible way; also, that the interpretation of the results be clearly presented, without misleading statements and without false conclusions. Laymen have difficulty enough in understanding the report of a water analysis. The analyst should seek to enlighten, not to obscure.

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* One grain per gallon equals 17.1 parts per million.

CHAPTER III

ODORS AND TASTES IN WATER SUPPLIES

The senses of taste and smell are closely related but distinct. Some substances, like salt, have a taste but no odor, and others, like vanilla, have a strong odor but no taste. Many of the so-called tastes are really odors, the gas or vapor given off by the substance tasted reaching the nose not only through the nostrils but also through the posterior nares. Thus an odor "tasted" is often stronger than an odor smelled.

Chemically pure water is free from both taste and odor. Water containing certain substances in solution, as salt and iron, may have a decided taste but no odor. Such taste-producing substances are found in mineral, brackish, or iron-bearing waters, but as a rule they are not offensive and seldom affect large bodies of water. Most of the bad tastes observed in drinking water are due not to inorganic but to organic substances in solution and suspension, or to microscopic organisms. These produce odors as well as tastes. In the following, the term "odor" is used to describe attributes of drinking water that may affect both the sense of smell and that of taste.

Water taken directly from the ground and used immediately is usually odorless. In certain sections of the country, deep well water has a sulphurous odor. Contaminated well water or water drawn from a swampy region may be somewhat moldy or unpleasant. Almost all surface waters have some odor. Many times it is too faint to be noticed by the ordinary consumer, though it can be detected by one whose sense of smell is carefully trained. On the other hand, the water in a pond may have so strong an odor that it is offensive several hundred feet away. Between these two extremes one meets with odors that vary in character and intensity, and are often the source of much annoyance and complaint.

The natural odors of waters are frequently intensified or changed in character by the use of chlorine as a disinfecting agent. Very disagreeable odors are sometimes encountered. The odor of free chlorine is in itself objectionable, but the odors and tastes produced by the combination of chlorine with certain organic substances in the water may give rise to even greater complaint. The use of chlorine for the hygienic protection of water supplies is of such great value and has become so wide-spread that the production of 'chlorination odors

and tastes" has added new interest to the study of odors and tastes in water supplies.

General Classification of Odors. — It is difficult to classify the odors of surface waters on a satisfactory basis, but they may be considered under three general headings:

1. Odors caused by organic matter other than living organisms.
2. Odors caused by living organisms.
3. Odors caused by chlorine or chlorine compounds and by the action of chlorine upon any of the preceding odor-producing agencies.

In reporting odors in water analyses it is customary to express the quality of the odor by a descriptive epithet as in Table 5, in which are also shown abbreviations used to simplify record keeping.

TABLE 5
QUALITY OF ODORS

<i>a</i> — aromatic	<i>m</i> — moldy
<i>c</i> — cucumber	<i>M</i> — musty
<i>Cl</i> — free chlorine, chlorinous	<i>N</i> — nasturtium
<i>d</i> — disagreeable	<i>o</i> — oily
<i>e</i> — earthy	<i>p</i> — pigpen
<i>f</i> — fishy	<i>s</i> — sweetish
<i>g</i> — grassy	<i>S</i> — hydrogen sulphide
<i>G</i> — geranium	<i>v</i> — vegetable
<i>I</i> — iodoform, medicinal	<i>V</i> — violets
<i>mm</i> — muskmelon	

The intensity of an odor may be indicated by using the prefixes *very faint*, *faint*, *distinct*, *decided*, *very strong*. A better method, however, is to use numerical prefixes, which may be approximately defined as shown in Table 6.

According to this method of reporting odors, for example, 3f indicates a *distinct fishy* odor and 2v a *faint vegetable* odor. Combinations of odors are also encountered, and it is often convenient to record such expressions as 3v + 2m, meaning a *distinct vegetable and faint moldy* odor. The reader will understand that these definitions are far from exact and that the intensity of odors that vary in quality cannot be well compared. A *faint fishy* odor, for example, might often attract more attention than a *distinct vegetable* odor.

Laboratory Determination of Odor. — In the laboratory the "cold odor" is observed by shaking a partly filled bottle of the water, removing the stopper, and quickly sampling the odor given off at the mouth of the bottle.

TABLE 6
INTENSITY OF ODORS

Numerical Value	Term	Approximate Definition
0	<i>None.</i>	No odor perceptible.
1	<i>Very faint.</i>	An odor that ordinarily would not be detected by the average consumer, but that could be detected in the laboratory by an experienced observer.
2	<i>Faint.</i>	An odor that the consumer might detect if his attention were called to it, but that would not attract attention otherwise.
3	<i>Distinct</i>	An odor that would be detected readily and that might cause the water to be regarded with disfavor.
4	<i>Decided.</i>	An odor that would force itself upon the attention and that might make the water unpalatable.
5	<i>Very Strong.</i>	An odor of such intensity that the water would be absolutely unfit to drink (a term to be used only in extreme cases).

Heating the water usually intensifies its odor. The "hot odor" is obtained by heating, to a point just short of boiling, a portion of the water in a tall beaker without a spout, covered with a watch glass. After partial cooling the cover is slipped aside and the odor quickly observed. A long deep breath should be taken at the instant of observation. Water that has a *faint* odor when cold may have a *distinct* odor when hot.

Odors Caused by Organic Matter.—The odors caused by organic matter other than living organisms may be classified as *vegetable* odors and "odors of decomposition." They vary in character in different waters and at different seasons. It is difficult to find terms that will describe them exactly, and different observers will seldom agree as to the most appropriate descriptive adjective. To one person the odor of a water sample may be *straw-like*, to another *swamp-like*, to another *peaty*. This is due to the fact that man's sense of smell is not well cultivated. In practice, therefore, it has become customary among analysts to use

the general term *vegetable* instead of the terms *straw-like*, *swamp-like*, *marshy*, *peaty*, *sweetish*.

Most of the *vegetable* odors are caused by colloidal vegetable matter. Brown-colored waters invariably have a *sweetish vegetable* odor, and the intensity of the odor varies almost directly with the depth of the color. Both color and odor are due to the presence of certain glucosides, of which tannin is an example, extracted from leaves, grasses, mosses, etc. In addition to an odor, these substances have a slight astringent taste. Colorless waters containing organic matter of other origin may have *vegetable* odors, but they are usually less *sweetish* and more *straw-like* or *peaty*. Akin to the *vegetable* odors are the *earthy* odors caused by finely divided particles of organic matter and clay. The two odors are often associated in the same sample.

Odors produced by the decomposition of organic matter in water are not uncommon. They are described, somewhat imperfectly, by such terms as *moldy*, *musty*, *unpleasant*, *disagreeable*, *offensive*. An *unpleasant* odor is produced when the vegetable matter in water begins to decay. It may be said to represent the first stages of decomposition. As decomposition progresses the *unpleasant* odors become *disagreeable*, and then *offensive*. It is seldom that the decomposition of vegetable matter in water produces odors worse than *decided unpleasant*. The *disagreeable* odors usually can be traced to decaying animal matter, and, as a rule, *offensive* odors are observed only in sewage or in grossly polluted water. The terms *moldy* and *musty* are more specific than the terms *unpleasant*, *disagreeable*, and *offensive*, but they are difficult to define. They are quite similar in character, but the *musty* odor is more intense and is usually, but not always, applied to sewage-polluted water. The *moldy* odor suggests a damp cellar, or perhaps a decaying tree trunk in a forest. The bacteriologist will recognize this odor as similar to that given off by certain bacteria growing on nutrient gelatine. The odors of decomposition naturally are associated at times with other classes of odors.

Odors Caused by Organisms. — The odors of drinking water that are due to the presence of living organisms are very important. They are common in occurrence, often offensive in nature, and may affect large bodies of water. It is only within recent years that these odors have been well understood, and even now there is much to be learned about the chemical nature of the odoriferous substances and their relation to the life of the organisms. At one time it was supposed that it was only by decay that the organisms became odoriferous. It is now well established that many living organisms have an odor that is natural and peculiar to them, just as a fresh rose or an onion has a natural and peculiar odor.

It has been found, also, that in most cases — and it may be true in all cases — the odor is produced by compounds analogous to the essential oils. Some of these oily compounds have been isolated by extraction with ether or gasoline. Odors due to them have been called “odors of growth” because the oils are produced during the growth of the organisms. In many genera the oil globules may be seen if the organisms are examined under a sufficiently high-powered lens. The oil globules are usually most numerous in the mature forms and are often particularly abundant just before sporulation or encystment. The production of the oil represents a storing up of energy. The odors have also been called “odors of disintegration,” because they are most noticeable when the breaking up of the organism scatters the oil globules through the water. It is sufficient, however, to call them the “natural odors” of the organisms, to distinguish them from the very different odors produced by decomposition.

Microscopic organisms are not commonly found in ground waters (except when stored in open reservoirs) or in rapidly flowing streams in sufficient abundance to cause trouble. It is in the quiescent waters of ponds, lakes, and reservoirs that they develop luxuriantly, and it is to the reservoir that one should look first when investigating the cause of an odor in a public water supply.

Odors of Littoral Organisms. — The organisms found on the sides of reservoirs include the flowering aquatic plants, the characeæ and the filamentous algæ, of the vegetable kingdom, and the fresh-water sponges, bryozoa, etc., of the animal kingdom. The effect that they exert on the odor of a water is difficult to determine because they are seldom found in a reservoir where floating microscopic organisms are wholly absent. In many cases where a peculiar odor in water has been attributed to some of these littoral growths, subsequent investigation has made it seem probable that the odor was really caused by limnetic organisms that had been overlooked in the first instance.

Speaking generally, it may be said that in reservoirs that are large and deep the organisms attached to the shores produce little or no effect on the odor of the water; and that in small, shallow reservoirs where aquatic vegetation is thick they do not impart any characteristic “natural” odor, but may produce a vegetable taste and a disagreeable odor due to decomposition.

Some of the littoral aquatic organisms, such as *Myriophyllum* and a number of the filamentous algæ, possess a natural odor that is strongly *vegetable* and, at times, almost *fishy*; but the odor is obtained only when the plants are crushed or when fragments are broken off and scattered through the water. Under ordinary conditions of growth in a reservoir

this does not happen, and therefore no odor is imparted to the water except through decomposition.

There are on record some apparent exceptions to the rule that the attached growths cause no odor. Hyatt has described a growth of *Meridion circulare* at the headwaters of the Croton River, in 1881, that was supposed to have affected the entire supply of New York City; Rafter has connected odors with *Hydrodictyon utriculatum* and other chlorophyceæ; Forbes has investigated a water supply in which a growth of *Chara* was thought to be the cause of a bad odor; Tighe has also reported a troublesome growth of *Chara* at Holyoke. Weston has stated that serious trouble was caused in Henderson, N. C., by an extensive growth of *Pectinatella*. All such cases, in which odors in water supplies have been attributed to certain littoral organisms, lack convincing scientific corroboration.

The author once examined a reservoir where a mass of *Melosira varians*, several feet thick, covered the slopes to a considerable depth. A severe storm tore away the fragile filaments, and masses of *Melosira* passed into the distribution pipes and caused a noticeable *vegetable* and *oily* odor in the water.

In connection with this relation of littoral organisms to odors in water supplies, some reference should be made to the *cucumber taste* that has been a frequent cause of complaint against the Boston water supply. In 1881 the trouble was very severe. The water had a decided odor of cucumbers, which was intensified at times to a *fish-oil* odor. Heating made the odor very strong and offensive. A noted analyst made an examination and concluded that the seat of the trouble was in Farm Pond — one of the sources of supply. This pond was so situated that all the water of the Sudbury system passed through it on its way to the city. Chemical analysis of the water and microscopical examination of the mud failed to reveal the cause of the odor. It was found, however, that fragments of fresh-water sponge (*Spongilla fluviatilis*) were constantly collecting on the screens and that these fragments had the *cucumber* odor. It was decided, therefore, that the fresh-water sponge was the cause of the odor. The conclusion was quite generally accepted and the report has been quoted extensively.

At that time some water analysts disagreed with this opinion. They claimed that the amount of sponge found in the pond was not sufficient to produce the odor. In the light of modern microscopical examinations we have come to believe that the dissenters were right and that the fresh-water sponge was not the cause of the *cucumber* odor. The author took masses of *Spongilla* and allowed them to rot in a small quantity

of water till the odor was unbearable. This water was then diluted with distilled water to see how large a mass of water the decayed sponge would affect. It was then found that with a dilution of 1 to 50,000 there was no perceptible odor. At this rate it would take a mass of sponge several feet thick over the entire bottom of Farm Pond to produce an odor as intense as that observed in 1881. Moreover, the odor produced by decaying sponge is not the *cucumber* odor, although similar to it.

There is good reason to believe that the *cucumber* odor observed in 1881 was due to *Synura*. One need not dispute the observation that the sponge that collected on the Farm Pond screens had the *cucumber* odor, for no doubt the sponge was covered with other organisms. It is not surprising, either, that *Synura* should have been overlooked in the water, because the organism disintegrates readily and a comparatively small number of colonies is able to produce considerable odor. The times of occurrence of the odor — namely, spring and autumn — are worth noting, as they correspond with the seasons when *Synura* grows best and is most commonly found.

In February, 1892, the cucumber taste again appeared in Boston water. This time it was definitely traced to *Synura* that was growing in the water just under the ice in Lake Cochituate. Since then it has reappeared at intervals in other parts of the supply — notably in Basins 3 and 6. It has been found that 5 or 10 colonies per cc. are sufficient to cause a perceptible odor.

Synura has often been the cause of bad odors in the Croton supply of New York, and it aroused nation-wide interest when it invaded the Catskill water supply in the winter of 1921-22.

Odors of Limnetic Organisms. — The floating microscopic organisms, or true plankton, are responsible for many of those peculiar nauseating odors that are the cause of complaint in so many public water supplies. In most, if not in all, cases, the odor is due to the presence of an oily substance elaborated by the organisms during growth. This has been proved by long-continued observations and experiments, during the course of which the following facts have been noted:

The odors referred to vary in character. They are difficult to describe, but can be readily identified. Particular odors are associated with particular organisms. If an organism is present in sufficient numbers its particular odor is observed; if it is not present in sufficient numbers its odor is not detected. With some exceptions, the intensity of the odor varies with the number of organisms present. If water that contains an organism possessing a natural odor is filtered through

paper, the odor of the filtered water* is much fainter than before, while the filter paper on which the organisms remain has a strong odor. If the organisms are concentrated by the Sedgwick-Rafter method, the concentrate has a decided taste and odor. If these organisms are placed in distilled water, the water acquires the odor of the original water. Thus, the relation between specific odors and specific organisms has been well established. Indeed, experienced observers are often able to tell the nature of the organisms present by observing the odor of the water in which they are found.

That the odors are not due to the decomposition of the organisms is proved by the character of the odors themselves and by the fact that the odors are not necessarily associated with the occurrence of large numbers of bacteria or with the presence of free ammonia or nitrites. Increasing numbers of bacteria and changes in the nitrogenous compounds always accompany the decomposition of microscopic organisms. Furthermore, when the organisms do decay, the odor of the water commonly changes in character.

The natural odor is produced by some substance within the cell wall of the organism, and when this substance is liberated the odor is more easily detected. The odor is intensified by heating, by mechanical agitation, by pressure, by change in the density of the water containing the organisms, and by chlorination. Many of the odor-producing organisms are very delicate. Heating breaks them up and drives off the odoriferous substances. The flow of water through the pipes of a distribution system is sufficient to cause the disintegration of many forms, and it is a matter of common observation that in such cases the odor of the water at the service taps is more pronounced than at the reservoir. If the density of water is increased by adding to it some substance, such as salt, the organisms may become distorted by osmotic pressure if not actually broken up. This causes an intensification of their odor. Increased pressure leads to the same result. The effects of chlorination will be discussed later.

The natural odor of microscopic organisms is due to some oily compound analogous to those substances, found in higher plants and animals, that give an odor to the peppermint and the herring, for example. The fact was noted long ago that the addition of salt to water affected with certain odors developed an oily flavor. Many of the tastes and odors caused by organisms are of a marked oily nature. In these organisms oil globules may be observed with the microscope. The

* In some cases the odoriferous substances from the organisms pass through the filter, and the disintegration of the organisms gives the filtered water an increased odor over the unfiltered water.

number of oil globules changes with the age and condition of the organisms, and the intensity of the odor varies with the number of oil globules present. Finally, oily substances have been extracted from the organisms and it has been found that they possess the same odor as that observed in the water containing them.

Algae as Local Nuisances. — Thus far in this chapter the algae have been considered from the standpoint of odors that are observed when the water is used for drinking. Algal odors are sometimes strong enough to be sensed also in the vicinity of reservoirs; in fact, in some cases, the odors have been wafted by the wind for distances of a quarter of a mile. The decay of littoral growths of filamentous algae may cause objectionable odors along the shore. The odors derived from the exposed bottoms of reservoirs, when the water has been drawn down, are familiar to all, but it is not generally known that such odors are largely due to algae.

Algae are sometimes driven inshore by the wind and stranded on beaches, where they decay and produce foul conditions. The "odor of the sea," which is so much loved, is due largely to stranded seaweed.

Odors of Essential Oils. — Experiments have shown that the amount of oil present in microscopic organisms is sufficient to account for the odors observed in drinking water. In these tests some of the familiar essential oils, such as oil of peppermint, oil of clove, and cod-liver oil, were diluted with distilled water, and the amount of the dilution at which the odor became unrecognizable was noted. Oil of peppermint was recognized when diluted 1 : 50,000,000; oil of clove, 1 : 8,000,000; cod-liver oil, 1 : 1,000,000. The odor of kerosene oil could not be detected when diluted 1 : 800,000. The amount of oil present in water containing a known number of organisms was estimated for comparison. It was found that in water containing 100 colonies of *Synura* per cc. the dilution of the *Synura* oil was 1 : 25,000,000; and that in water with 50,000 *Asterionella* per cc. the dilution was only 1 : 2,000,000. Thus, the production of the odor by the oil is quite within the range of possibility. An interesting fact brought out by the experiments was that the odor of the oils varied with different degrees of dilution, not only in intensity but also in character. On one occasion, seven people out of ten who were asked to observe the odor of very highly diluted kerosene oil declared that it smelled like "perfumery." This variation of the character of the odor with its intensity is important, as it accounts for the different descriptions of the same odor in a water supply at different times and by different people.

The nature of the odoriferous oils or oily substances is not well known. Calkins, who, with the aid of gasoline and ether, isolated the odoriferous

principle of Uroglenopsis,* describes it as being similar to the essential oils. It was non-volatile at the temperature of boiling water. Jackson and Ellms extracted, with gasoline, a similar substance from *Anabæna*. On standing, it oxidized and became resinous. It contained needle-like crystals. Experiments by the author have shown that the oils of *Asterionella* and *Mallomonas* are quite similar in character.

Most, if not all, of the organisms produce oil during their growth, to a greater or less degree. In many cases it is quite odorless. Water is often without odor even when large numbers of organisms are present. This is either because the organisms have not produced oil, or because the oil is odorless. Sometimes water rich in organisms has an oily flavor with no distinctive odor. This is true in the case of some species of *Melosira*. Many organisms impart a vegetable and oily taste, without a distinctive odor. This is true of *Synedra pulchella* and *Stephanodiscus*. There are, moreover, microscopic organisms that produce oils that have a distinctive odor, but that occur in drinking water in such small numbers that the odor is not detected. The organisms that have a distinctive odor and that are found in large numbers are comparatively few. Not more than twenty-five have been recorded and only about half a dozen have given serious trouble. More extended observations may lengthen this list.

The distinctive odors produced by these organisms may be grouped around three general terms — *aromatic*, *grassy*, and *fishy* — and for convenience they may be tabulated as in Table 7.

Aromatic Odors. — The aromatic odors are due chiefly to the diatomaceæ. The strongest odor is that produced by *Asterionella*. The character of this odor changes with its intensity. When few organisms are present the water may have an undefinable *aromatic* odor; as they increase the odor resembles that of a *rose geranium*; when they are very abundant the odor becomes *fishy* and *nauseating*. The other diatoms given in the table produce the aromatic odor only when present in very large numbers. There are two protozoa that have an aromatic odor. The odor of *Cryptomonas* is *sweetish* and resembles that of the *violet*. The odor of *Mallomonas* is similar to that of *Cryptomonas*, but when strong it becomes *fishy*.

Grassy Odors. — The *grassy* odors are produced largely by the cyanophyceæ. *Anabæna* is the most important organism of this class. There are several species that have slightly different odors. The *grassy* odor is usually accompanied by a *moldy* odor, which is probably due to decomposition, as this organism decays rapidly. When strong the odor of *Anabæna* much resembles *raw green-corn*, or even a *nasturtium* stem.

* *Uroglenopsis americana*, formerly called *Uroglena americana*.

TABLE 7
ODORS OF PARTICULAR ORGANISMS

Group	Organism	Natural Odor
AROMATIC ODOR	DIATOMACEÆ	
	Asterionella	Aromatic — geranium — fishy.
	Cyclotella	Faintly aromatic.
	Diatoma	Faintly aromatic.
	Meridion	Aromatic.
	Tabellaria	Aromatic — geranium — fishy.
	Synedra	Earthy.
	PROTOZOA	
	Cryptomonas	Candied violets.
	Mallomonas	Aromatic — violets — fishy.
GRASSY ODOR	CYANOPHYCEÆ	
	Anabæna	Grassy and moldy — green-corn — nasturtium — pigpen.
	Aphanizomenon	Grassy — nasturtium — pigpen.
	Clathrocystis	Sweet, grassy.
	Cœlosphærium	Sweet, grassy.
	Cylindrospermum	Grassy.
	Rivularia	Grassy and moldy.
	CHLOROPHYCEÆ	
	Dictyosphærium	Grassy — nasturtium — fishy.
FISHY ODOR	DIATOMACEÆ	
	Asterionella	Slightly fishy (in large numbers).
	Tabellaria	Slightly fishy (in large numbers).
	CHLOROPHYCEÆ	
	Dictyosphærium	Faintly fishy (through chlorination).
	Eudorina	Faintly fishy.
	Pandorina	Faintly fishy.
	Volvox	Fishy.
	PROTOZOA	
	Bursaria	Irish moss — salt marsh — fishy.
	Ceratium	Vile stench (rusty brown color).
	Dinobryon	Fishy, like rockweed.
	Glenodinium	Fishy.
	Mallomonas	Fishy (in large numbers).
	Peridinium	Fishy, like clam-shells.
	Synura	Ripe cucumbers — muskmelon — bitter and spicy taste.
	Uroglenopsis	Fishy and oily — cod-liver oil.

NOTE: Additions to this table after Dr. F. E. Hale.

The following organisms have caused trouble other than that connected with characteristic odor: *Fragilaria*, *Melosira*, *Navicula*, *Cœlastrum*, *Cladophora*, *Tribonema*, *Spirogyra*, *Oscillatoria*, *Beggiatoa*, *Crenothrix*, *Leptothrix*, *Chara*.

In extreme cases it is best characterized as a *pigpen* odor. The prevailing odor, however, is *grassy*, i.e., the odor of freshly cut grass. The other blue-green algæ have odors that may be called *grassy*, but they are less distinctive than the odor of *Anabæna*. One of the green algæ, *Dictyosphaerium*, also produces a *grassy* odor.

Fishy Odors.—The *fishy* odors are the most disagreeable of any observed in drinking water. The odor produced by *Uroglenopsis* is perhaps the worst. It is quite common. Water rich in *Uroglenopsis* has an odor not unlike that of *cod-liver oil*. The odor of *Synura* is almost as bad and almost as common. It resembles that of a *ripe cucumber*. *Synura* also has a *distinct bitter* and *spicy* taste. This taste “stays in the mouth” and is most noticeable at the back part of the tongue. *Glenodinium* and *Peridinium* both produce *fishy* odors. That of the latter somewhat resembles *clam-shells*. *Dinobryon* has a *fishy* odor and suggests *sea-weed*. The odor of *Bursaria* is described as being like that of *Irish moss*. It also reminds one of a *salt marsh*. With certain degrees of dilution, some other protozoa have the *salt-marsh* odor, reminding one of the sea. *Fishy* odors are produced by *Volvox*, *Eudorina*, and *Pandorina*. These chlorophyceæ are sometimes classed with the protozoa, so that it may be said in a general way that the *fishy* odors are produced by microscopic organisms belonging to the animal kingdom. When present in large numbers, some diatoms (*Asterionella* and *Tabellaria*) also produce a *fishy* odor.

Odors of Plankton Decomposition.—Some of the microscopic organisms have distinctive odors of decomposition. The cyanophyceæ when decaying give a *pigpen* odor. *Beggiatoa* and some species of *Chara* give the odor of *sulphureted hydrogen*. All the odors given off by the decomposition of microscopic organisms are offensive. They are particularly so when the organisms contain a high percentage of nitrogen. Jackson and Ellms, in an interesting study of the decomposition of *Anabæna circinalis*, found that this organism contained 9.66 per cent of nitrogen. They found that the *pigpen* odor was due “to the breaking down of highly organized compounds of sulphur and phosphorus and to the presence of this high percentage of nitrogen. The gas given off during decomposition was found to have the following composition:

Marsh gas	0.8%
Carbon dioxide	1.5%
Oxygen	2.9%
Nitrogen	12.4%
Hydrogen	82.4%
	<hr/>
	100.0%

The gas that remained dissolved in the water containing the *Anabæna* was practically all CO_2 and represented a large percentage of the total gas produced."

Chlorination Odors. — The odors caused by the use of chlorine or chlorine compounds as disinfecting agents may be attributed to one or more of the following causes: 1. Excess or "free" chlorine. 2. Substitution compounds of chlorine with organic matter of animal or vegetable origin. 3. Substitution compounds of chlorine with phenoloid or comparable substances. 4. Destruction of organisms resulting in the release of aromatic substances which in some cases unite with chlorine.

Excess or Free Chlorine. — The *chlorinous* taste, as the name implies, is imparted to water by an excess of chlorine, i.e., by an amount greater than that necessary to enter into combination with the chlorine-absorbing matter in the water. Since waters vary greatly in composition, very small amounts produce *chlorinous* tastes in some waters — well waters and filtered waters for example — whereas relatively large quantities are absorbed by other waters. *Chlorinous* tastes and odors have been absent in tap water containing much organic matter when dosed with sufficient amounts of chlorine to show a "residual" of 0.8 to 1.0 part per million of chlorine at the point of application. Certain spring waters containing little organic matter and much bicarbonate of lime, on the other hand, have produced a slight odor of chlorine at cold-water taps and considerable odor at hot-water taps when a "residual" of 0.1 p.p.m. of chlorine was recorded. "Residual chlorine" is commonly defined as the "free" chlorine present in a dosed water after 5 to 15 minutes' contact at 20° C. The difference between the initial dose and the residual is the chlorine demand of the water. A strong correlation exists between the chlorine demand and the oxygen demand of different waters. A residual of 0.2 p.p.m. is now commonly required in the disinfection of drinking water as a factor of safety for the destruction of bacteria after the initial chlorine demand of the water has been satisfied. This practice takes no recognition of the fact that the ultimate chlorine demand of waters differs widely.

Chlorinous tastes were quite common in the early days of chlorination, but with modern methods of mechanical application and laboratory control complaints due to this type of odor should be very few and confined principally to those waters which in their raw state are relatively clean from the physical standpoint but which are subject to bacterial contamination.

Substitution Compounds of Chlorine with Phenoloid or Comparable Substances. — The most disagreeable taste produced in water by the use of chlorine is the *iodoform* or *medicinal* taste* which occurs when

phenoloid or comparable substances are present. Extremely small concentrations of these substances, such as 1 in 1000 million (0.001 p.p.m.) are able to impart an *iodoform* taste to certain waters, and, once established, the taste will persist.

The phenols, cresols, and other comparable bodies find their way into water chiefly by the discharge of industrial wastes from coke by-products plants, city gas plants, producer gas plants, and similar industries. Phenoloid substances are also contained in the atmosphere near cities and industries, and pure chlorinated water exposed to their influence may acquire a *decided medicinal* taste, especially when rain, fog or mist precipitates the polluting substances on the water surface.

Abundant experiences with *iodoform* tastes are on record. The obnoxious tastes in the Milwaukee water supply, traced clearly to the operation of coal-distillation plants discharging wastes into the river and lake from which the city's water supply is taken, are well known. *Medicinal* tastes have been reported in chlorinated water carried by freshly dipped pipe or stored in newly coated standpipes and elevated tanks. The effect of atmospheric pollution has been investigated, more particularly by Adams in England.

Pure water seems to be more sensitive to taste production when chlorinated than water containing organic matter. Thus, at London, treatment of the Thames river water has been readily accomplished, while great difficulty has been experienced with the New River which contains much less organic matter. Undoubtedly there are other factors, such as temperature, that influence taste production.

Destruction of Microscopic Organisms. — When used in sufficient concentration chlorine will destroy algæ as well as bacteria. The addition of chlorine to water, therefore, often results in the death and disintegration of microscopic organisms and the release of the aromatic odor-producing oils. These remain free or combine with the chlorine to form very noticeable or intensified odors. Concentrations of organisms that are not objectionable in unchlorinated water become decidedly so when chlorine is added. Thus 300 standard units of *Synura* have been considered necessary to cause the well-known cucumber taste in unchlorinated water, whereas the New York experience of 1921 shows that concentrations of *Synura* as low as 25 units may cause trouble in a chlorinated supply. Dr. Hale reports, in fact, that it is possible to taste *Synura* in the chlorinated water usually about one week before it can be discerned under the microscope in concentrations of one or two standard units. Similar experiences have been recorded for other plankton organisms.

Fortunately the discovery was recently made that chlorine can serve

as a cure for troubles of this type that it may cause. When used in excess it not only destroys microscopic organisms but also reacts with the liberated oils to form odorless compounds. This dosage of chlorine is known as superchlorination and presents a very useful method for the control of odors and tastes. Superchlorination is discussed more fully in Chapter XIII.

Besides these chlorine odors, water supplies sometimes become affected with other "chemical odors" — such as those of carbolic acid, creosote, tar, etc. They can be traced usually to some pollution by manufacturing waste, though a vigorous decomposition of organic matter has been known to give an odor resembling carbolic acid. Similar odors are sometimes caused by the coating on the inside of new distribution pipes or the paint in stand pipes and elevated tanks. Tastes in water supplies are not necessarily associated with pollution, the growth of microscopic organisms, or the use of disinfectants. Dissolved mineral salts can be tasted in many normal waters. The concentration capable of detection by the individual depends upon his sensitiveness.

Occurrence of Odors in Massachusetts Water Supplies. — The extent to which water supplies are afflicted with odors is well shown by the following table based on Massachusetts experience. The predominance of objectionable odors in surface water supplies is here made evident. The great number of people who must drink water that has a *distinct* or *decided* odor during a considerable part of the year points to the importance of studies in the microscopy of drinking water. Almost three million users of surface water in Massachusetts consume water that has a noticeable odor about a third of the time.

The quality of the odors in Massachusetts supplies has been investigated by Calkins who states that in 1404 samples from surface water supplies in Massachusetts odors were observed as shown in Table 9.

The intensity of these odors was not stated. Many of them probably were not strong enough to cause complaint. This is shown by the weekly odor records kept in the laboratory of the Metropolitan Water Works during the years 1905 to 1920 (Table 10).

The assumption was made in compiling Table 10 that all odors are objectionable, whatever their character; that *very faint* and *faint* odors are noticeable; and that *distinct* and *decided* odors are objectionable. It is seen that in the Metropolitan District the odor of the water is strong enough to cause complaint for two or three weeks each year.

It must not be inferred from the foregoing tables that Massachusetts is more afflicted in her water supplies than other sections of the country. The same troubles are observed almost everywhere. It is only because data for Massachusetts supplies are more readily[†] available than for

TABLE 8
INTENSITY OF ODORS IN MASSACHUSETTS WATER SUPPLIES
(Average of Monthly Analyses 1910 to 1919)

Source of Supply	No of Cities and Towns	Population, 1920	Per Cent of Samples of Stated Odor				
			None	Very Faint	Faint	Distinct	Decided and Strong
<i>Surface Water</i>	82	2,865,747	8.7	19.8	39.5	27.8	4.1
Boston, Metropolitan..	18	1,206,849	1.3	20.2	57.3	20.5	0.7
Worcester.....		179,754	0.2	9.1	56.4	32.7	1.6
Springfield.....		129,563	23.6	14.3	32.6	25.8	3.7
New Bedford.....		121,217	0.0	2.5	36.0	47.8	13.7
Fall River.....		120,485	0.0	7.6	64.7	25.2	2.5
Cambridge.....		109,694	0.0	0.7	30.8	58.7	9.8
Lynn.....		99,148	0.0	1.7	30.0	56.8	11.5
Lawrence.....		94,270	0.0	84.2	15.8	0.0	0.0
<i>Ground Water</i>	75	583,386	91.3	4.1	3.3	1.1	0.2
Lowell.....		112,759	67.3	26.7	5.6	0.4	0.0
Newton.....		46,054	95.3	1.5	1.6	1.6	0.0
Brookline.....		37,748	95.2	3.7	1.1	0.0	0.0
<i>Surface and Ground Water</i>	31	254,361	44.3	16.0	26.0	11.5	2.1
Chicopee.....		36,214	41.4	12.9	30.3	14.8	0.6
North Adams.....		22,282	8.5	38.8	44.3	8.4	0.0
Framingham.....		17,033	93.7	3.5	2.1	0.7	0.0

Compiled from: Whipple, G. C. A Rating of the Qualities of the Water Supplies of Massachusetts, Journ. N. E. W. W. Assn., XXXVI, p. 40.

TABLE 9
QUALITY OF ODORS IN MASSACHUSETTS WATER SUPPLIES

Odor	Per Cent of Samples Affected	Odor	Per Cent of Samples Affected
None.....	20	Fishy.....	3
Vegetable.....	26	Moldy.....	10
Sweetish.....	7	Disagreeable.....	6
Aromatic.....	6	Offensive.....	7
Grassy.....	15		

others that attention has been drawn to them. Microscopic organisms are widely distributed both in this country and abroad. Wherever they are found in abundance they will inevitably affect the odor of the water.

TABLE 10
ODORS IN METROPOLITAN WATER SUPPLY, BOSTON, 1905 to 1920
Per Cent of Time that Stated Odor was Observed

	Practically No Odor	Odor too Faint to Attract Attention	Odor No- ticeable but not Enough to Cause Complaint	Odor Strong Enough to Cause Some Complaint	Odor Strong Enough to Cause Gen- eral Com- plaint
Wachusett Reservoir.....	4.4	85.8	5.8	3.2	0.8
Sudbury Reservoir.....	1.5	73.3	15.2	9.0	1.0
Framingham Reservoir, No. 2	1.0	80.7	10.0	8.1	0.2
Lake Cochituate*.....	0.4	38.8	29.5	23.6	7.7
Chestnut Hill Reservoir.....	3.0	84.4	8.2	3.5	0.9
Spot Pond Reservoir.....	3.0	78.2	13.2	4.8	0.6
Tap, 180 Boylston Street.....	1.9	84.7	8.0	4.9	0.5
Tap, Ashburton Place.....	1.7	84.5	7.4	5.6	0.8

* Held as reserve supply and seldom used.

Value of Pure Water. — In his little book entitled “The Value of Pure Water,” the author has attempted to express in terms of money† the value to a community of a supply of clean water over a water that is unattractive by reason of color, turbidity, and the presence of algæ. Since this treatise is no longer in print the following paragraphs are taken from it.

The analytical determinations which relate to the general attractiveness of a water are those of taste, odor, color, turbidity, and sediment. As these quantities increase in amount, the water becomes less attractive for drinking purposes, until finally a point is reached where people refuse to drink it. In order to use these results in a practical way, it is necessary to combine them so as to obtain a single value for the physical characteristics or, as they say abroad, for the “organoleptic” quality of the water. An attempt has been made by the author to obtain what may be termed an æsthetic rating of the water, and the result is shown in Fig. 2.

This diagram, it should be said, is based almost entirely upon estimates and very little upon statistical data. It rests upon the assumption that people differ in their sensibilities or their æsthetic feelings as

† Prices are as of 1907.

to the use of water. Some persons are much more fastidious than others in regard to what they drink. A water which would be shunned by one person, even though he were thirsty, might be taken by another with apparent relish. As a rule, people are more fastidious about the odor of water and the amount of coarse sediment which it contains than they are about its color and turbidity. This is perhaps natural, as a bad odor suggests decay, and decay is instinctively repugnant. Often, however, people do not discriminate between odors which are due to decomposition and those which are not. Habit and association have much to do with a person's views as to the attractiveness of water. In

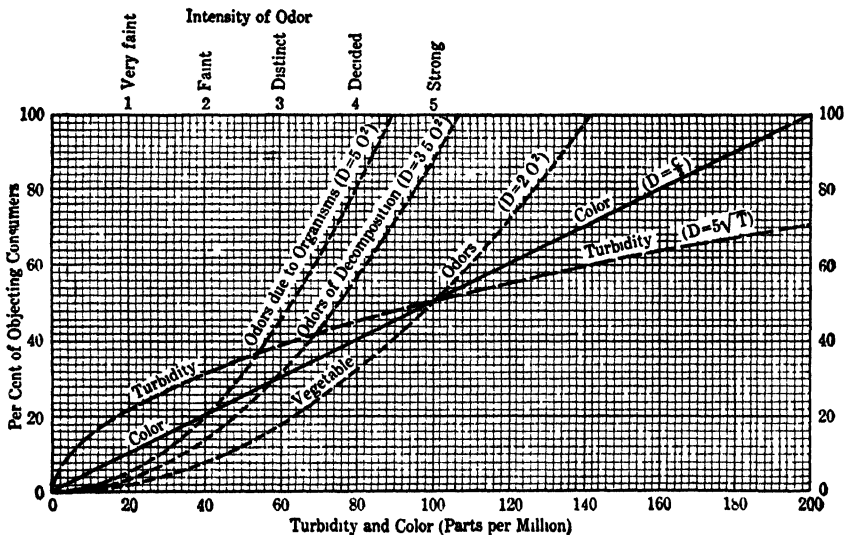


FIG. 2. — *Æsthetic Deficiency of Water.*

The "*Æsthetic Deficiency*" is found by adding together the "*Per Cents of Objecting Consumers*" for Color, Turbidity and Odor.

New England, where the clear trout brooks run with what Thoreau called "meadow tea," few people object to a moderate amount of color, while they do object to a water which is very turbid. In the Middle West, where all the streams are muddy, it is the unknown colored waters which are disliked. People who are accustomed to well water object to both color and turbidity. With most people a fine turbidity, such as is produced by minute clay particles, is less a subject of complaint than an equal turbidity produced by comparatively coarse sediment. In the diagram an attempt has been made to reconcile these different points of view, so as to put them, as well as may be, on the same footing. In this connection several series of comparisons were made.* Turbid waters were viewed by a group of Western people, who made some com-

* Acknowledgments are due to Mr. J. W. Ellms of Cleveland, Ohio, and Mr. Andrew Mayer, Jr., of Brooklyn, N. Y.

parisons with colored and turbid waters, while colored waters were viewed by a group of students in New York, and *vice versa*.

The abscissæ of the diagram represent turbidity, color, and odor, as given in the ordinary water analysis. The ordinates represent the "per cent of objecting consumers." By this is meant the proportion of the water takers who would ordinarily choose not to drink the water because of the quality indicated by the curve, or who would buy spring water, or bottled water, rather than use the public supply, if they could afford to do so. This number would increase, of course, as the general attractiveness of the water decreased. From the curves one may calculate what may be called the *æsthetic deficiency* of the water by adding together the per cents of objecting consumers for color, turbidity, and odor. If the *æsthetic deficiency* equals 100, it indicates that the water is of such a character that every one would object to it, and figures in excess of 100 only emphasize its objectionable character.

It will be seen from the diagram that when the color of water is less than 20, or the turbidity less than 5, only one person in ten would object to it, but when the turbidity or color is 100, one-half of the people would object to it. It may be thought that this proportion is too low, but it must be remembered that colored waters are invariably accompanied by a vegetable odor and often by a slight turbidity, and that it is the sum of the several quantities which determines the *æsthetic* rating.

Experience has shown that objection to color varies directly with its amount; consequently this curve has been plotted from the equation, $p_c = 0.5 c$, i.e., a straight line, where p_c stands for the per cent of objecting consumers, and c for the color.

In the case of turbidity, however, small amounts count for more, relatively, than larger amounts. The equation for the turbidity curve has been taken, therefore, as $p_t = 5 \sqrt{t}$, where t stands for the turbidity.

With odor, however, the opposite condition prevails; faint odors count for little, but distinct and decided odors cause much more complaint. Consequently, the per cent of objecting consumers has been made to vary as the square of the intensity of the odor expressed according to the standard numerical scale. The quality of the odor makes quite as much difference as its intensity, and for that reason three curves have been plotted, one representing vegetable or pondy odors (O_v), one representing odors due to decomposition (O_d), and one representing the aromatic, grassy and fishy odors due to microscopic organisms (O_o). These curves are plotted from the following equations:

$$\begin{aligned} p_o &= 2 O_o^2, \\ p_o &= 3.5 O_d^2, \\ p_o &= 5 O_v^2, \end{aligned}$$

in which O_o , O_d , and O_v stand for the intensity of the three groups of odors mentioned.

These curves represent somewhat imperfectly our present ideas as to the relative effects of color, turbidity, and odor; and on further study they are likely to be considerably modified.

It is a well-known fact that in cities which are supplied with water which is not attractive for drinking purposes, large quantities of spring

water and distilled water are sold, and that consumers go to much expense in the purchase of house filters in order to improve the quality of the water furnished by the city mains. It is fair to assume that in any community the amount of money expended for bottled water and house filters will vary in a general way, according to the attractiveness of the water, although there is no doubt that the presence of typhoid fever in the community, or the fear that the water is contaminated, will greatly increase the use of auxiliary supplies for drinking. For purposes of calculation it may be assumed that the diagram just described represents this tendency to use vended waters, and that each "objecting consumer" would go to the expense of buying spring water or putting in a house filter, if he could afford it. It may be argued, also, that the poor consumer who may be unable to do this is as much entitled to satisfactory water as is the well-to-do consumer.

From a study of price lists of spring waters sold in New York and other cities, it has been found that the ordinary wholesale price of spring water is seldom more than 10 cents a gallon. In some places it is as low as 1 cent. The average is about 5 cents. To filter water through house filters costs less, but generally it is less satisfactory.

As a convenient figure for calculation, and as a most conservative one for general use, a cost of 1 cent per gallon to the ordinary consumer for an auxiliary supply of drinking water (either spring water or well-filtered water) has been taken. In cities where the cost of procuring and distributing bottled water exceeds 1 cent per gallon, as it does in such a city as New York for example, this should be taken into account in making local use of the data. For the illustrative purposes of the present study, and for general comparisons, the figure mentioned will serve as a satisfactory basis. The average person drinks about 1.5 quarts of liquid per day, of which one-half may be assumed to be water, the rest being tea, coffee, etc. Therefore one-fifth cent per capita daily may be taken as a reasonable figure for the cost of an auxiliary supply. If the entire population used such a supply, and if the daily consumption of the public water supply were 100 gallons per capita, then one-fifth cent per hundred gallons, or \$20 per million gallons, would represent the loss to the consumers due to an imperfect water-supply which had an æsthetic deficiency of 100. If the æsthetic deficiency were less than 100, say 37, then the loss to the consumer would be $\frac{37}{100}$ of \$20, or \$7.40 per million gallons. In other words, the figure for the æsthetic deficiency divided by 5 gives the financial depreciation of the water supply in dollars per million gallons, or

$$D = 20 \frac{p_c + p_t + p_o}{100}.$$

Example: Suppose the turbidity of a water is 3, its color 65, and its odor 2f (that is, faint fishy), because of the presence of microscopic organisms; then

$$D = 20 \frac{12 + 32 + 20}{100} = \$12.80;$$

that is, the depreciation of the water, because of its unsatisfactory physical qualities, amounts to \$12.80 per million gallons.

TABLE 11

DEPRECIATION DUE TO ODOR

Values of D for different values of O_v , O_d , and O_o in the formula

$$D = \frac{20 (2 O_v^2 + 3.5 O_d^2 + 5 O_o^2)}{100}$$

Dollars per million gallons

	Odor	Vegetable Odor (O_v)	Odor of Decomposition (O_d)	Odor Due to Organisms (O_o)
0.....	None	0.0	0.0	0.0
1.....	Very faint	0.4	0.7	1.0
2.....	Faint	1.6	2.8	4.0
3.....	Distinct	3.6	6.3	9.0
4.....	Decided	6.4	11.2	16.0
5.....	Strong	10.0	17.5	25.0

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CHAPTER IV

COLLECTION OF SAMPLES

The first question that confronts the aquatic microscopist is the collection of material for examination. This requires the selection of suitable sampling apparatus. The choice of equipment must naturally depend upon the object and scope of the studies to be made. Some of the collecting devices used by water biologists are extremely simple; others possess complex mechanisms. Ordinarily the water analyst deals only with bottle samples. These permit a quantitative estimate of the various organisms present in the sample and furnish the microscopic information required in routine sanitary analysis. Special investigations of surface and ground waters that are to serve as sources of water supply and the biological control of lakes and rivers, however, often necessitate studies that call for more elaborate apparatus, such as plankton nets, pumps, scoops, and dredges. Much time and labor can be saved by the proper choice and use of collecting devices.

COLLECTION OF PLANKTON SAMPLES

Generally speaking, there are two types of plankton-sampling apparatus: one by which the aquatic organisms are collected suspended in the water in which they have their habitat; the other by which they are strained from the water during collection, thus yielding a concentrated sample or catch. Whenever possible, the analyst himself should supervise the collection of samples. If he attempts to draw inferences from analyses of water about the collection of which he knows nothing, he may do so at the risk of his reputation.

Bottle Samples. — In collecting water samples in bottles, all types of microscopic life are included, and this method of sampling is generally to be preferred to others in the routine sanitary analysis of water. The quantity of water required for microscopical examinations depends upon the nature of the water and the object of the analysis. For routine microscopical examinations forming part of a sanitary water analysis, one quart is sufficient. When a chemical analysis is to be made as well as the microscopic, two quarts or a gallon is necessary. For researches into the composition of the plankton, the study of the flora

and fauna of ponds, lakes, and rivers, and other limnological or rheological investigations, larger quantities are sometimes required.

Surface Samples. — The collection of a sample of water from or near the surface of a stream, pond, or lake, or from a service tap or pump, requires no special collecting apparatus. Any glass bottle of good quality can be used after thorough cleansing. In taking a sample from a service tap or pump, the water should be allowed to run for several minutes before the bottle is filled so that a fresh sample will be drawn. The bottle should not be filled completely; a small air space should be left below the stopper of the bottle to allow for expansion.

In collecting a sample from a stream, care must be exercised to exclude floating material and to prevent the stirring up of bottom deposits. By pointing the mouth of the bottle downstream, surface débris can often be kept out. This method, however, should not be used when the sample is to undergo chemical or bacteriological analysis, as the water, before flowing into the container, comes into contact with the hands of the collector and may become contaminated or altered in composition. The collection of a sample from a pond requires careful selection of the sampling point. The plankton is moved about by wind and wave action; it is not found dispersed uniformly through the water, and it is difficult to obtain a sample truly representative of the horizontal and vertical distribution of the organisms. It may be necessary to take several samples or to prepare a composite one. Floating scum should be excluded as in the case of other surface waters, unless special information is desired upon scum growths. It is well to note the nature of the littoral plant life in the vicinity of the sampling station, as records of plant growths are sometimes of value in the interpretation of analyses.

Deep Samples. — In the study of water supplies it often becomes necessary to collect samples at different depths below the surface of the body of water investigated. The simplest deep-sampling device consists of a weighted bottle that is lowered to any desired depth before the stopper is removed. After the bottle has become filled, air bubbles cease to rise to the surface and thus indicate to the collector that the bottle may be safely hauled in. But little water will be displaced from a full bottle while it is being drawn to the surface.

When special equipment is not available it is possible to devise a number of simple rigs for deep sampling. Some of these are illustrated in Fig. 3. Fig. 3a shows the bottle attached to the cord by means of two half-hitches about the neck and body. A large stone can serve as the weight. The stopper is attached to an auxiliary cord. When samples are collected at appreciable depths, the auxiliary cord is apt to become entangled with the main cord and prevent withdrawal of the

stopper. In order to avoid the use of two cords the bottle may be rigged as shown in Fig. 3b. Here the main cord is attached to the stopper by means of a slip knot. The bottle itself is held by a short

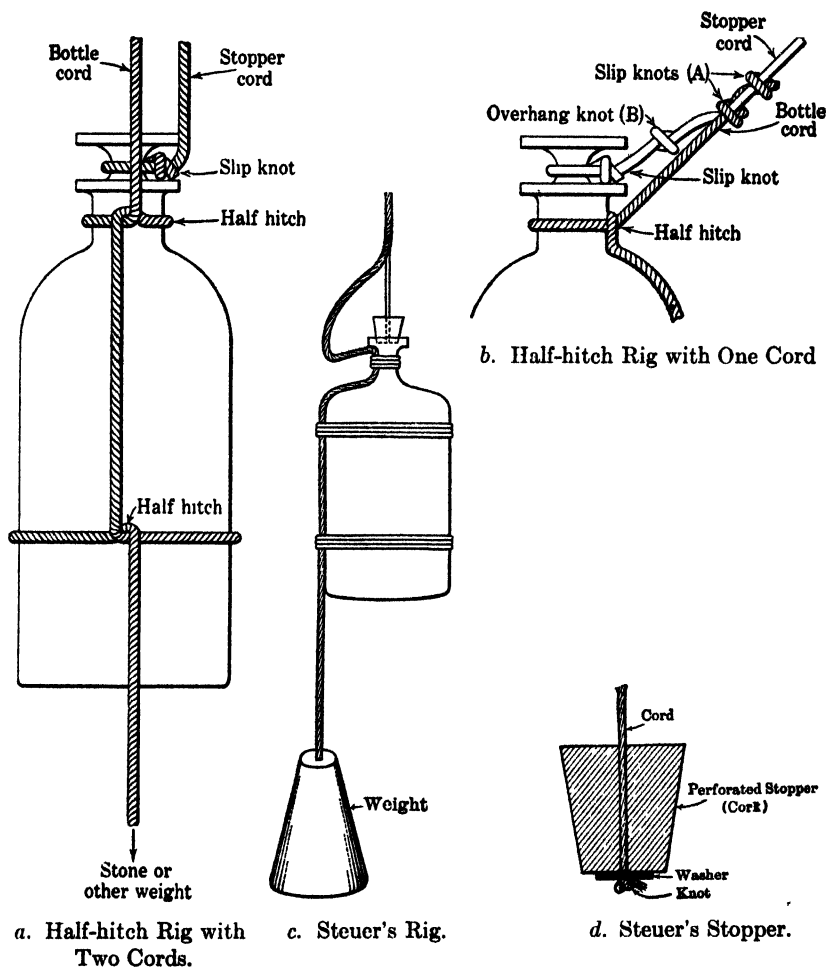


FIG. 3. — Simple Deep Sample Collectors.

second cord in a manner similar to that shown in Fig. 3a. One or more slip knots (A) fasten the auxiliary cord to the main cord and provide sufficient friction to leave the stopper cord slack while the bottle is being lowered. When the bottle reaches the desired depth a sudden jerk on the line permits the stopper cord to slip sufficiently through the friction knots to remove the stopper from the mouth of the bottle;

«knot (*B*) then catches on the friction knots and prevents further slipping of the cord. A rig used by Steuer is shown in Figs. 3*c* and 3*d*. With this arrangement the bottle is cast into the water on a slack line, a length of cord reaching to the desired depth having been played out beforehand. When the bottle reaches the predetermined depth, the cord straightens, becomes taut, and the stopper is pulled out. Should the stopper remain in place, a sharp jerk on the cord will dislodge it.

The cord on which the sampling outfit is lowered and raised can be graduated by marking the measuring units with paint, colored yarn, or adhesive tape. To insure reasonably accurate measurements the cord, before being calibrated, should be wetted and, while weighted, shrunk. The best material to use is a good grade of sash cord which comes in sizes of 3/16 inch to 3/8 inch, known commercially as No. 6 to No. 12 cord. For depths exceeding one hundred feet, tiller cord can be employed. This contains a bronze wire cable which insures adequate strength and prevents undue stretching or shrinking.

Whipple's Collecting Device. — A sampling device developed under the author's direction in the Laboratory of Sanitary Engineering of the Harvard Engineering School is shown in Fig. 4.

The apparatus is essentially a brass cage in which the glass sampling bottle is enclosed. The cage is composed of a bent brass rod, *A*, attached to a pan, *B*, partly filled with lead. This gives the apparatus a weight of about 25 pounds. Attached to the wire frame are straps *C*, *D*, and *E* which hold the bottle in place. Straps *C* and *D* are hinged at *F* and *G* and clamp to strap *E* by means of knurled screws, *H* and *I*. The entire frame is supported by a spring, *K*, joined to the sinking-cord, *L*. A flexible cord, *M*, extends from the top of the spring to the stopper of the bottle. The length of this cord and the length and stiffness of the spring are so adjusted that when the apparatus is suspended in the water by the sinking-rope the cord is just a little slack. In this condition it is lowered to the depth at which the bottle is to be filled. A sudden jerk given to the rope stretches the spring and produces sufficient pull on the cord, *M*, to remove the stopper. As a precaution against a possible loss of the apparatus through breaking of the spring, a safety cord, not shown in the figure, extends through the helix connecting the sinking-rope, *L*, directly to the frame, *A*. This safety cord, which is always a little slack, is furthermore of a length that will prevent too great a stretching of the spring. For use at great depths, additional weight may be given the apparatus by lead disks, *N*, weighing about 10 pounds, snapped to the bottom of the cage.

When the downward water pressure on the stopper overbalances the resistance and the weight in water of the apparatus, it becomes difficult

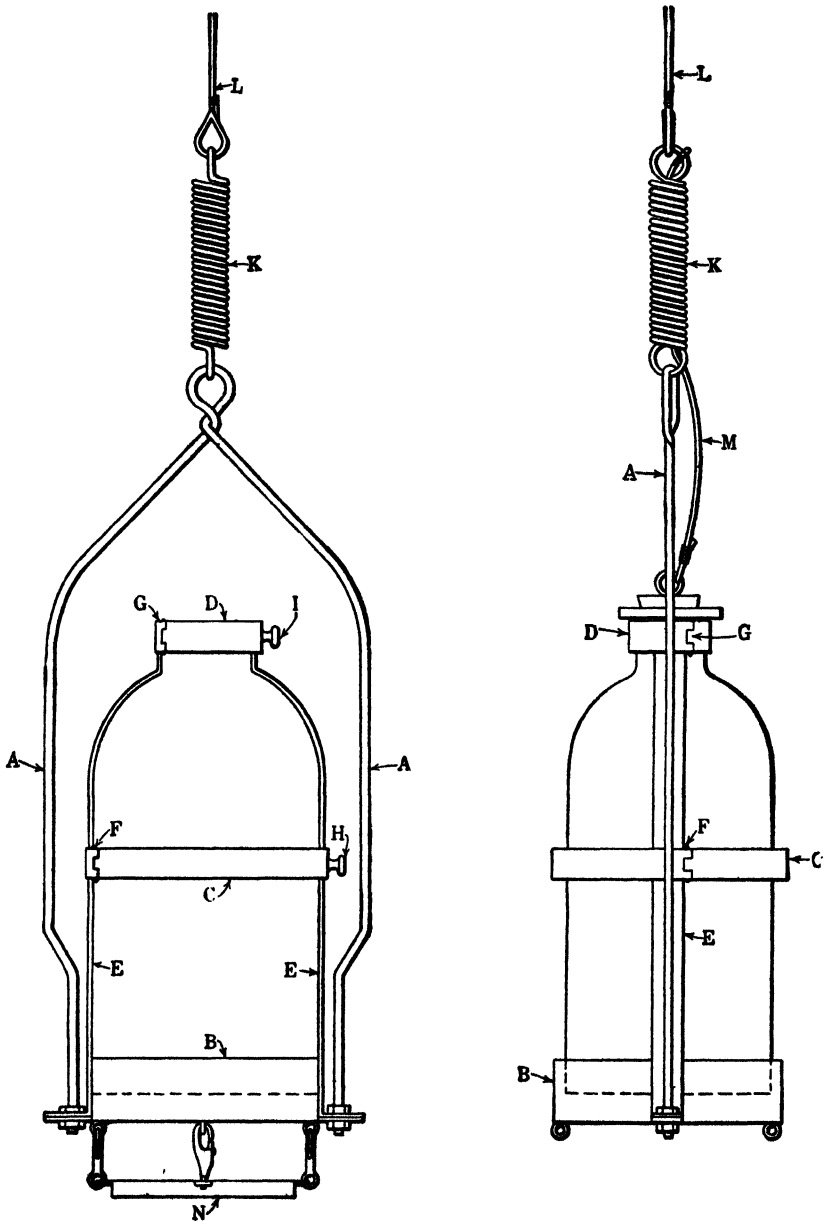


FIG. 4. — Whipple's Collecting Device.

to dislodge the stopper. The smaller the stopper the more easily is it removed in deep water. The size of the aperture through which the water enters the bottle may be reduced by passing a piece of brass tubing through a rubber stopper and closing the tube at the top with a small rubber stopper or a brass plug ground to fit. A spring may also be used to break off the sealed tip of a glass tube inserted in the stopper. A good stopper mechanism was devised by Richard H. Eurich, while a student of sanitary engineering in Harvard University.

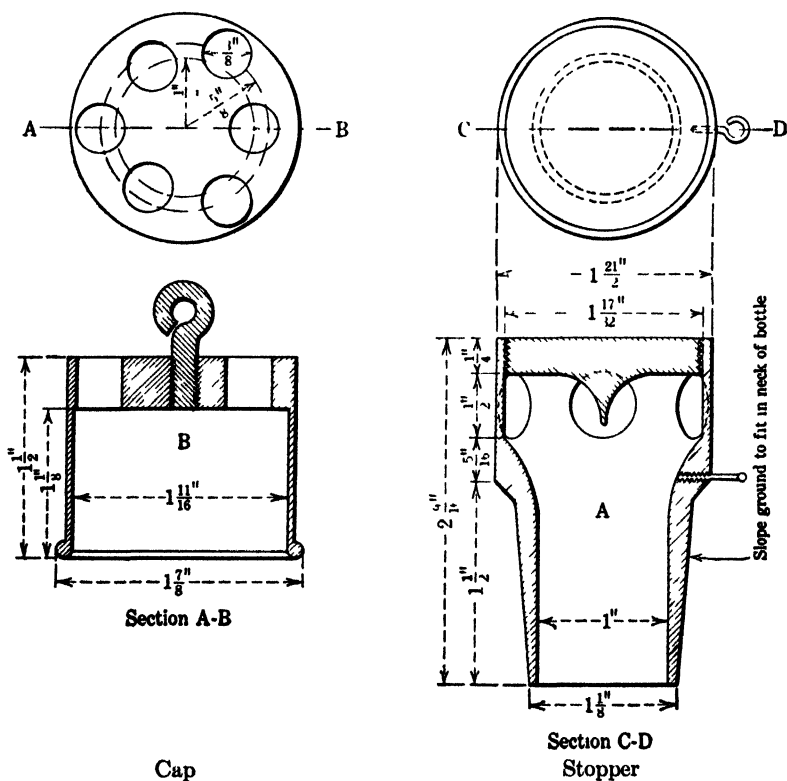


FIG. 5. — Eurich's Stopper for Water Sampling Bottle.

Eurich's Stopper for Water-Sampling Bottle. — Eurich used a balanced valve for admitting water to a bottle. The valve, or stopper, is shown in Figs. 5 and 6. It is constructed of brass or other suitable non-corroding metal and consists of two parts, the mouth piece, A, and the cap, B. The mouth piece is ground to fit into the neck of the bottle and contains four ports through which the water enters the bottle. The cap seats easily over the mouth piece and closes the ports. The

top of the cap is perforated in order to offset the vertical downward water pressure upon it. The releasing line is attached to the cap, which is pulled off when the line is jerked. This allows the water to enter the bottle through the ports. The apparatus is hauled to the surface without closing the ports, since experience has shown that the entrance of water on the way up is negligible. Numerous other sampling devices are in use.

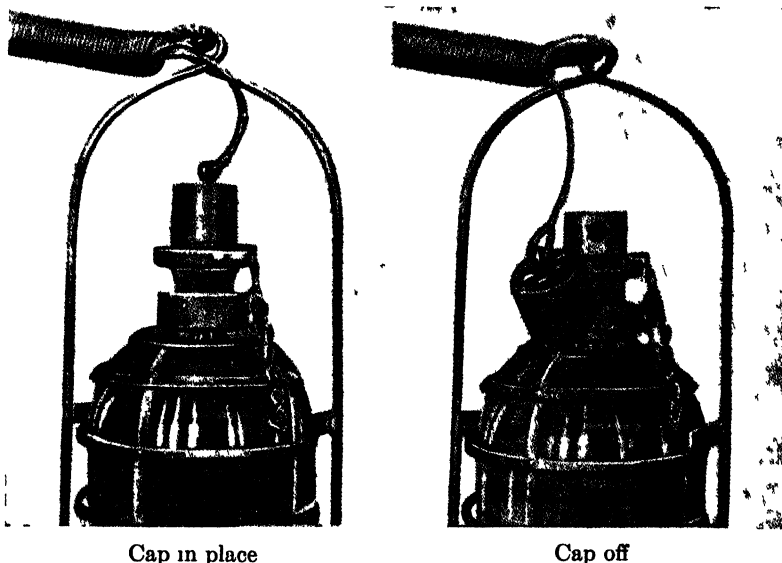


FIG. 6. — Collecting Bottle Equipped with Eurich's Stopper.

Hale's Sampling Bottle. — Dr. Frank E. Hale has adapted, for use in obtaining samples for microscopic analysis, a sampling device commonly employed in the collection of water samples for the determination of their dissolved oxygen or other gaseous content. The apparatus is shown in Fig. 7. The principles underlying its operation are discussed in Chapter VIII.

This type of sampling bottle is especially useful when an integrated sample of water is to be taken, i.e., when the sample is to be a composite of the vertical layers of water. Since the head of water on the bottle is independent of depth, uniform motion of the apparatus will collect nearly equal volumes of water in equal intervals of time. The varying compressibility of the air in the bottles interferes somewhat with the uniformity of sampling.

The other types of "dissolved oxygen" samplers described in Chapter VIII can also be used successfully for the collection of plankton. The

combined sampling device shown in Fig. 56 is of value in collecting samples through the ice cover of lakes and streams.

Kemmerer-Foerst Water Bottle. — The sampling devices so far mentioned all make use of glass bottles. These are easily broken, especially when the bottom of the stream or lake is rocky. A metallic collector devised by Dr. G. I. Kemmerer of the United States Bureau of Fish-

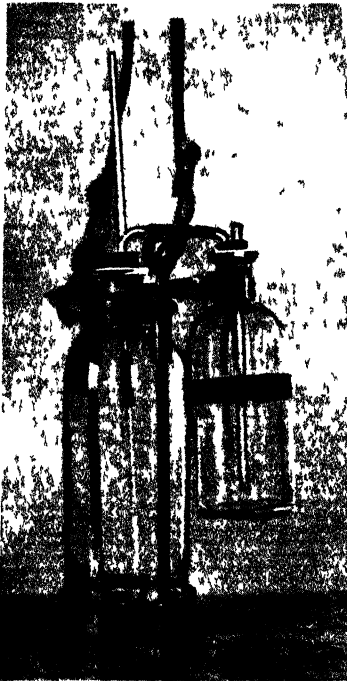


FIG 7. — Hale's Sampling Apparatus

eries, and modified by Foerst, is shown in Fig. 8. It consists of a brass tube closed by rubber stoppers which are activated by a messenger sent down the suspension line.

A somewhat similar collecting device used in deep-sea exploration and known as the "Scottish" water bottle is made by Negretti and Zambra, London, England. Kofoed has used a water or plankton trap consisting of a tube 6 feet long and 4 inches in diameter, provided with a suitable valve. S. M. Ellsworth has devised a simple method of collecting an integrated sample representative of part or all of the water in a vertical column. His apparatus consists of a garden hose provided at its lower end with a check valve opening inward. Water passes into the hose while it is being slowly lowered, and the check valve effectively seals the opening when the hose is withdrawn. The water collected in the

hose is transferred to a sampling bottle.

Plankton Nets. — The plankton net was first used by Hensen in 1882 in his investigation of the Baltic, North Sea, and Atlantic Ocean. As shown in Fig. 9 it consists of three parts, an inlet or guard cone, a straining cone, and a concentrating bucket. The guard cone conducts the water into the net through which it is strained. The microörganisms are retained on the filtering material and are then washed into the bucket whence they are easily transferred into a phial or collecting bottle.

Construction of Plankton Nets. — Birge and Juday, in their extensive study of Lake Mendota, used two sizes of nets. Their small net consists of a truncated canvas cone secured to a framework of 4 mm. brass

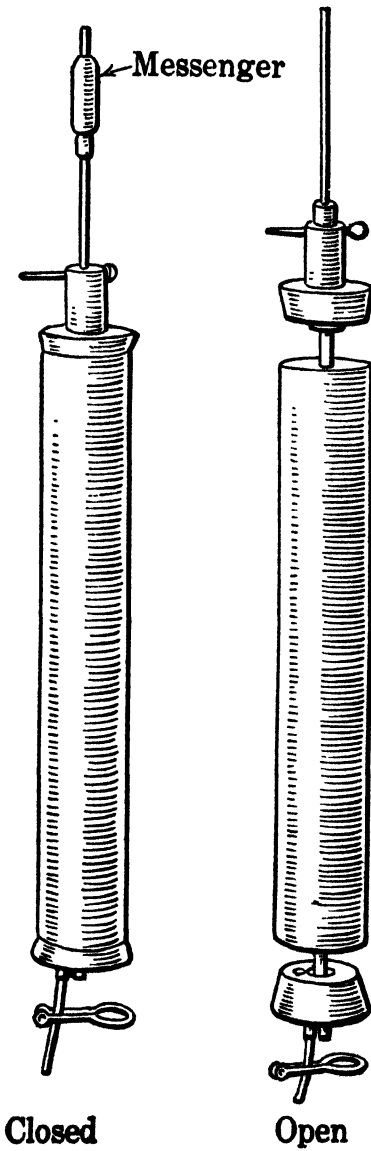
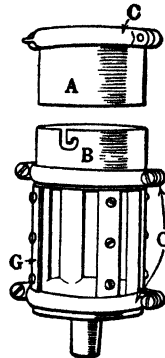


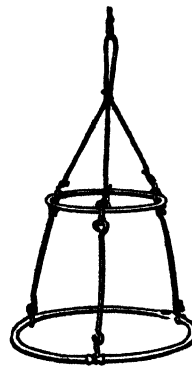
FIG. 8. — Kemmerer-Forrest Water Bottle. *After Birge.*



Assembly



Plankton Bucket



Framework

FIG. 9. — Plankton Net. *Bucket and Framework after Juday.*

wire (Fig. 9b). The upper ring is 12 cm. in diameter, the lower 18 cm. The two rings are held apart by three connecting wires. The straining cone of the net is 30 cm. long and is made of No. 20 (new No. 25) silk bolting cloth which when thoroughly shrunk possesses over 6000 meshes per square centimeter. The area of the openings varies from 0.001 to 0.003 sq. mm. To the bottom of the cone a bucket 5.2 cm. in diameter is secured (Fig. 9c). The bucket consists of a head piece, *A*, attached directly to the bolting-cloth strainer, and the bucket proper, *B*. The head and walls are of brass telescope tubing of such size that the head fits easily but snugly into the top of the bucket. The head is guyed to the canvas cone by means of three cords slightly less than 30 cm. long, which take the strain from the bolting cloth. The bucket has four windows covered with bolting cloth. The side piece, *G*, and two clamps, *C*, secure it in place and allow ease of removal. The clamp *C'* secures the bolting-cloth cone to the head piece. Two bayonet clutches fasten the bucket to the head piece. The bucket has a slightly conical bottom in the middle of which there is an outlet closed by a removable plug or screw. The whole net is suspended by three cords looped together and fastened to the rope by which the apparatus is raised or lowered.

The large net used by Birge and Juday is similar in shape to the small one. The opening of the truncated cone is 25 cm. in diameter and the lower ring 30 cm. The canvas cone is 33 cm. long, and the bolting-cloth cone 70 cm. The same size of plankton bucket is employed as with the smaller net. For use in connection with sanitary water analysis, the author prefers the smaller net.

Reighard's net, used at Lake St. Clair, was 3 feet in length, 2 feet in maximum diameter, with an opening 16 inches in diameter. In Allen's net the canvas guard net is replaced by a brass cone with a surface area of 100 sq. cm. A haul of 1 meter then theoretically filters 100 liters of water.

Operation of Plankton Net. — There are two methods of operating plankton nets. In the older method the net serves both as the collecting device and the straining or concentrating medium; in the newer one it serves as the straining medium only.

When the net is both to collect and to concentrate the organisms, it is lowered to the bottom or to any other desired depth and is then drawn to the surface, the velocity of its ascent being noted. A number of tests have shown that the water passes from the outside through the net while it is being lowered except when it is operated from a boat in rough weather. When the water is not smooth the net moves up and down with the motion of the boat and an error is introduced. On the

way up the net theoretically filters a column of water, the cross-section of which is that of the opening of the guard net and the height of which is equal to the distance through which the net is drawn. Actually, the net does not strain the whole column of water through which it passes, as a portion of the water is forced aside. In order to obtain the volume of plankton in the column traversed, it is therefore necessary to correct the values obtained. The ratio of the catch that would be obtained if all the water passed through the net, to the actual catch is called the "net coefficient." This coefficient varies for each net and for different velocities of ascent through the water. It also varies with the amount of clogging and the age of the net. New nets should be shrunk. Velocities less than 3 feet per second are commonly used and coefficients as low as 1.2 have been obtained. This means that the catch obtained must be multiplied by 1.2 in order to approach closely the true number or volume of organisms in the column of water traversed by the net. The efficiency of the net is the reciprocal of the net coefficient expressed in per cent. It is, therefore, the per cent of organisms caught. A coefficient of 1.2 thus represents an efficiency of 83 per cent. It is necessary to know the coefficient for each net at different velocities and to correct the results for each haul for the particular velocity used. A mathematical expression can be derived to establish the theoretical net coefficient. A nearly uniform velocity of ascent can be secured by providing the net with a float. After the net has been lowered to the desired depth, the weight and line are detached by a messenger and the net rises by itself to the surface.

When the net reaches the surface, it is allowed to drain. A stream of water played on the outside of the net detaches the organisms from the bolting cloth and washes them down into the bucket. The bucket is then unclamped and the collected material is transferred to a small bottle for transportation to the laboratory.

In order to permit the sampling of water strata lying at varying depths below the surface, Birge has devised a closing net in which a messenger, travelling down the rope holding the net, operates a release which allows the guard net to fall to one side, thus closing the net at any depth. He has also used a plankton trap consisting of a box-like structure, with movable top and bottom, surmounting the net in place of the guard cone. This trap gives accurate results and permits by comparison the determination of the coefficient of plankton nets.

Other modifications of the plankton net have substituted a 4-ounce bottle or other solid container for the bucket or have replaced the bucket by a cotton disk similar to that described later in this chapter. If it is desired to collect organisms in weeds or along the bottom where the

bolting-cloth net might be injured, it is possible to replace the cloth by a stronger material of loose weave. The results obtained naturally depend upon the type of cloth used.

Instead of operating the net vertically, it is sometimes better to haul the net obliquely through the water. This increases the length of haul and permits the collection of a composite horizontal and vertical sample. The oblique-haul method is employed particularly in rivers when the net is commonly hauled across the channel. It requires the use of a guide rope along which the net travels.

Of late, the net has been used more frequently as a straining medium



FIG. 10. A piece of bolting cloth No. 20 with plankton organisms drawn between the meshes to show relative sizes.

Upper row, left mesh: *Gymnodinium*, beneath *Amphidinium rotundatum* and *Exuviaella ballica*, right *Pouchetia parva*; middle mesh: *Prorocentrum micans* and *Rhynchomonas marina*; right mesh: *Nitzschia sigmaella*, *Achnanthes pulchra*, *Halteria rubra*, *Nitzschia closterium*. Middle row, left mesh: *Tintinnopsis nana*, *Tintinnus stenotrupis*, *Ocyrrhis phaeocysticola*; middle mesh: chain of small *Chaetoceras* species, above it on the left *Thalassiosira nana* and *saturni*, on the right *Carteria*; right mesh: chain of large *Chaetoceras* species (*Chat. didymum*), *Tintinnopsis beroides*. Lower row, left mesh: *Rhodomonas ballica*, *Distaplia speculum*; middle mesh: *Strombidium caudatum* (?), *Meringosphaera mediterranea*, *Amoeba*; right mesh: *Coccolithophora wallichi*, beneath on the left *Pontosphaera huxleyi*, on the right *Coccolithophora leptopora*, above on the right *Chrysomonadine* without shell, at the very bottom *Rhabdosphaera claviger*. $\times 110$. After Lohmann.

only. Known amounts of water are pumped into it from different depths. The pumps are calibrated for the length of hose used, and the quantity of water strained may be determined approximately by counting the number of strokes made with the pump for each catch. A more accurate method of procedure is to place the net in a calibrated pail. The water is pumped into the net very slowly and passes through it into the pail. Care must be taken to prevent the jet of water from striking the bolting cloth directly. The advantage of this method over the older one is that the amounts of water filtered can be accurately determined.

Net-Plankton and Nannoplankton.

— The plankton net does not retain all of the organisms contained in the water which is strained through it. Many of them pass through the meshes of the net, and only those with dimensions larger than the net opening are caught. The plankton retained are therefore designated "net-plankton"; those that escape, "nannoplankton" (i. e., dwarf

plankton). They have also been called "mesoplankton" and "microplankton," respectively. The size of the nannoplankton in relation

to the meshes of No. 20 bolting cloth is shown in Fig. 10. Lohmann has applied the term nannoplankton arbitrarily to those organisms whose maximum diameter does not exceed 25 microns.

There is no constant relationship between the number of nannoplankton and net-plankton found in water. Authorities agree, however, that nannoplankton are far more abundant than net-plankton and play an important part in aquatic biology.

According to Juday, the nannoplankton consist of such forms as rhizopods, flagellates, ciliates, rarely a rotifer, and various forms of algæ. The rhizopods are represented by an occasional *Ceratium*, *Peridinium*, and *Euglena*; the ciliates, by such forms as *Paramecium*, *Halteria*, *Coleps*, and *Vorticella*. The algæ belong to two general groups, namely, those that are so small that they are regularly lost by the net, and those that are lost only by accident. To the former belong such forms as *Ankistrodesmus*, *Oöcystis*, *Chodatella*, *Sphærocystis*, and some species of *Coclesphærium* and *Microcystis*. Those lost accidentally are young individuals or colonies, individuals so small that they are readily lost through the meshes of the net when the catch is being concentrated in the bucket, elongated or rod-shaped individuals that pass through when they strike the net endwise, and fragments of larger colonies. To this group belong forms such as *Anabæna*, *Aphanizomenon*, *Melosira*, *Stephanodiscus*, *Cyclotella*, *Tabellaria*, and *Fragilaria*.

The loss of the nannoplankton in using the plankton-net method must be kept in mind. The method of concentrating the catch should always be noted, as different methods, such as net, Sedgwick-Rafter, filter-paper, and centrifugal methods, yield different proportions of the total organisms present in water. In sanitary biology the plankton-net method is a valuable adjunct to the bottle-sampling methods discussed above. Its limitations, however, must be recognized.

Plankton Filters. — Filtration through sand will retain the smaller plankton better than straining through bolting cloth. As shown in Chapter V, sand filtration is the basic method of concentrating samples in the laboratory for routine sanitary analysis. When many samples are to be collected it is sometimes advisable, in order to reduce the bulk of the samples that must be transported, to perform the concentration in the field. While the common laboratory apparatus which is described later can be used for this purpose, less fragile equipment is more desirable.

Sling Filter. A simple sand-filtration device patterned after the Sedgwick-Rafter funnel has been developed by the author. It consists of a metal funnel of known volume to which is attached, by means of a

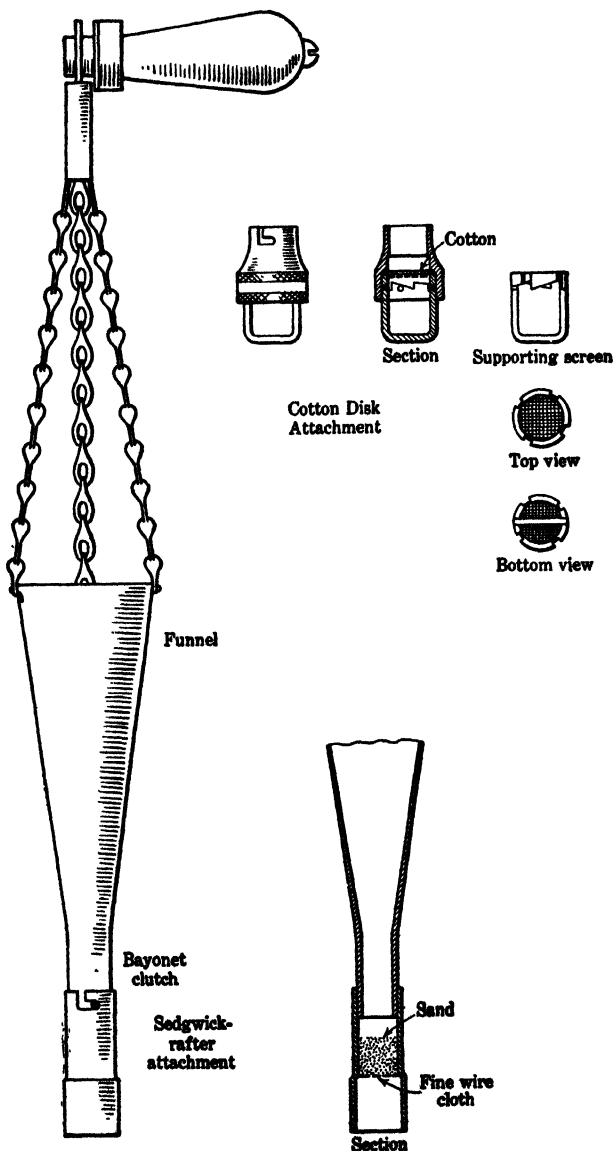


FIG. 11. — Sling Filter with Attachments for Use in the Field with the Sedgwick-Rafter Method and Cotton Disk Method.

bayonet clutch, a small cup which supports a column of sand on fine wire gauze. The water to be concentrated is collected in a bottle, poured into the funnel, and filtered through the sand. Filtration is hastened by swinging the funnel around the handle axis. The construction of the sling filter is shown in Fig. 11. Its operation is the same as that of the Sedgwick-Rafter funnel (see p. 91). Surface samples can be collected by the funnel directly. Deep samples must either be pumped into the sling filter or poured from deep-sample bottles.

The Cotton-Disk Filter. — An interesting and valuable method of keeping a permanent visual record of the amount of suspended organic

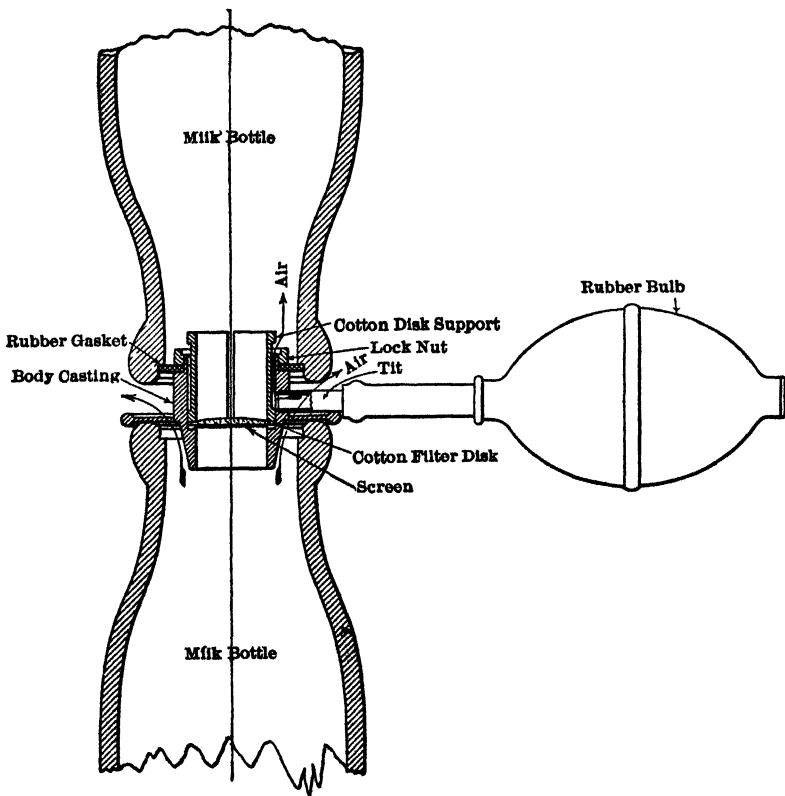


FIG. 12. — Wizard Sediment Tester.

matter in water, or showing the relative distribution of suspended materials in a body of water, and of obtaining rapid results in algal control tests is that of filtering a large volume of water through a thin sheet, or plug, of cotton. This can be done in the field as well as in the

laboratory. Although this method is not one of great accuracy from the standpoint of the analyst, it is an excellent one for showing to the eye the variation in algal growths in public water supplies. The best method of filtration is that which was originally devised for the determination of dirt in milk, known as the "Wizard Sediment Tester," made by the Creamery Package Manufacturing Company of Boston, Mass. The filtering medium is a thin wafer of cotton about an inch in diameter, which is held between two supports in a cap attached to a glass milk bottle. The water to be filtered is placed in the bottle and allowed to flow out through the cotton. Filtration is hastened by increasing the air pressure within the bottle by the use of a hand bulb. In order to filter a sufficient volume of water the bottle has to be filled several times. The cotton disks dry readily on blotting paper and can be mounted on cards to form a permanent record. The relative abundance of algæ or other suspended matter in the water is shown by the discoloration of the cotton. Figure 12 shows the apparatus and Plate B illustrates the variations in the appearance of the cotton after filtering five-quart samples of Cambridge water on different days.

The author has been strongly impressed with the practical value of this method and believes that it should be used more generally by water-works superintendents. It forms a useful addition to the field equipment for testing water, is inexpensive, and appeals to the layman as presenting visual evidence of the condition of the water.

As shown in Fig. 11, the author has devised a cotton-disk attachment for the sling filter. The disk takes the place of the sand as a filtering medium. The cotton rests on wire gauze and is held in place by a clamp. Filtration is hastened as with the sand.

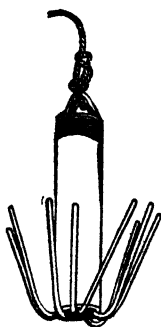


FIG. 13. — Pieter's Plant Grapple.
From Ward and Whipple.

COLLECTION OF LARGE AQUATIC ORGANISMS AND BOTTOM SEDIMENTS

Many of the organisms of interest to sanitarians live in or attached to the mud deposits on the bottom of rivers, lakes, and reservoirs. The occurrence of some of these organisms is of especial importance in the interpretation of stream-pollution studies; others aid in the explanation of chemical as well as biological analyses.

Plant Grapple. — Larger aquatic plants that grow in deep water and are not readily reached by hand can be gathered by means of a plant grapple. Pieter's plant grapple, Fig. 13, is made by passing several

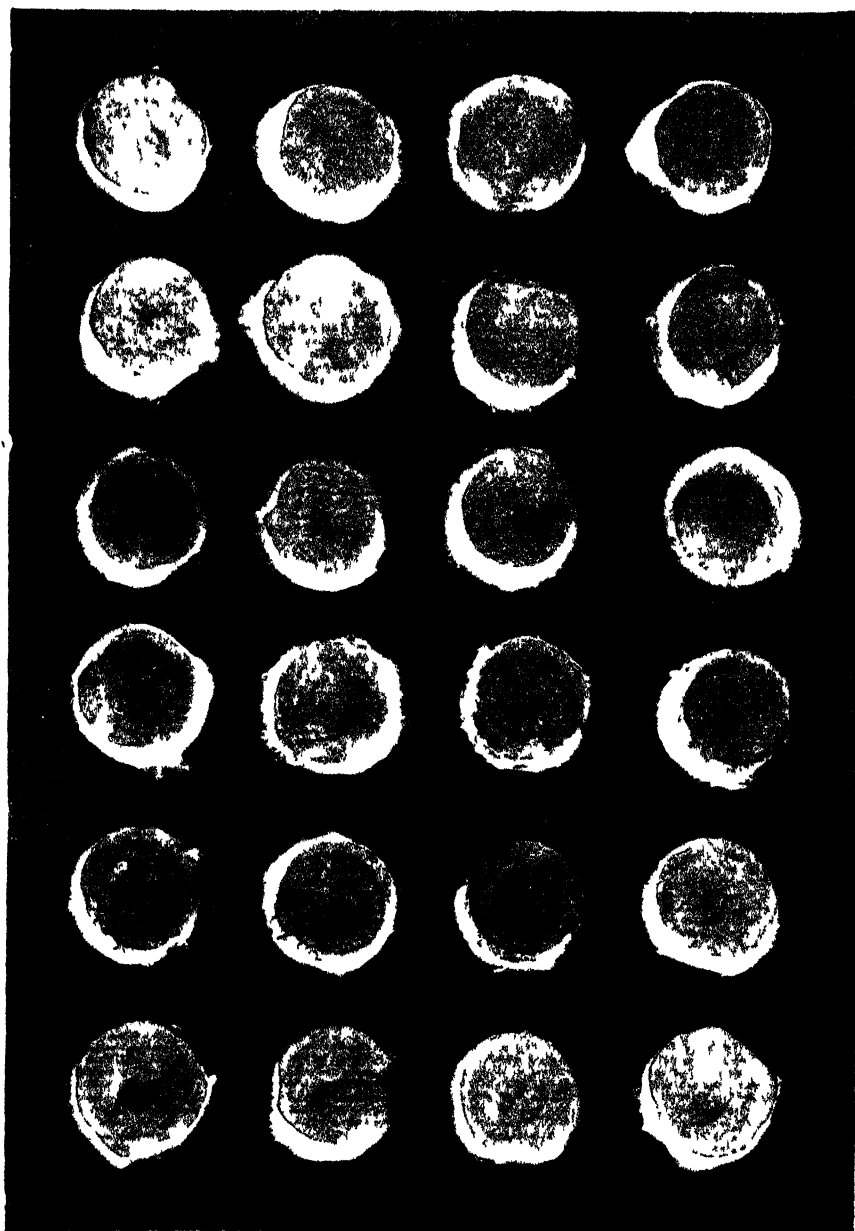


PLATE B

Sediment (chiefly algæ) from Cambridge Tap Water Collected on Cotton Discs at
Different Times during the Autumn of 1913

bent steel bars in the form of a double S through a piece of pipe. The pipe can be weighted with lead to make it heavier. A rope is fastened through the loop of the bars projecting through the top of the pipe.

Scoop and Dredge. — Bottom sediments can be sampled by means of scoops or dredges. A simple scoop used by the United States Public Health Service is illustrated in Fig. 14. The scoop is mushroom-shaped and collects about 3 liters of sediment. It is cast into the water and dragged from 5 to 15 feet along the bottom by an attached rope. A representative sample of about 200 cc. is selected from the material collected, placed in a half-liter bottle, and taken to the laboratory for examination. When necessary or convenient, the sample can be freed from mud in the field by wash-

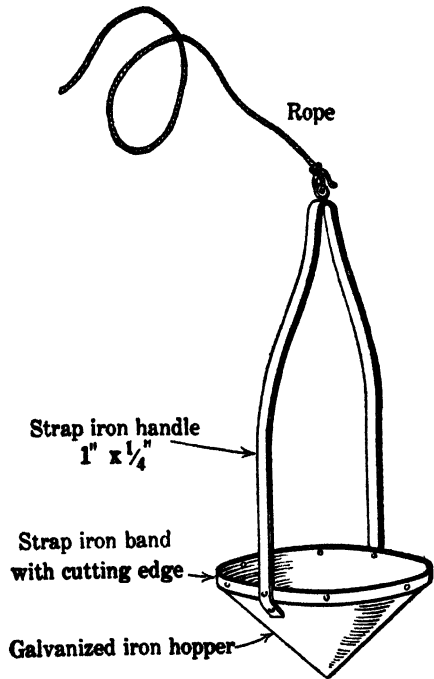


FIG. 14. — Mud Sampler. After Purdy. *U. S. Hygienic Laboratory Bulletin 104.*

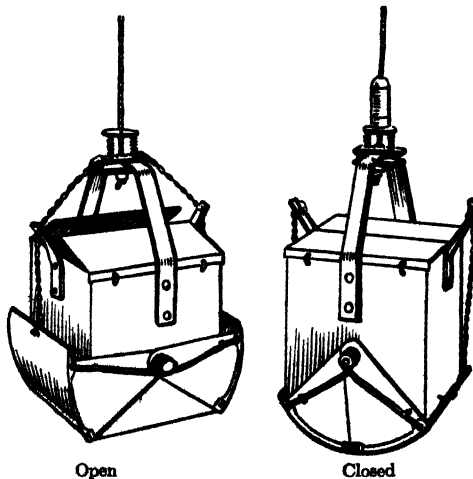


FIG. 15. — Ekman Dredge.

ing it through a 30-mesh sieve. The material retained on the sieve is then placed in a bottle of water.

Limnologists have made extensive use of the Ekman dredge shown in Fig. 15. This dredge operates similarly to a clam-shell bucket dredge. After it has been lowered to the bottom, the jaws are released by means of a messenger, and scoop up sufficient mud to fill about two-thirds of the bucket.^a This dredge oper-

ates successfully only on mud bottoms, as the springs working the

jaws are not strong enough to force them into sand, past sticks or rocks. Numerous other forms of dredges and scoops have been devised.

A small sample of bottom sediment can be obtained by forcing a pipe into the mud. The core thus obtained is removed after the pipe has been withdrawn. Pipes are particularly useful on hard or boulder-covered bottoms.

A dredge designed for the capture of organisms that live on the bottom but do not burrow into the mud is shown in Fig. 16. This dredge is similar in construction to the plankton net but is rectangular in section.

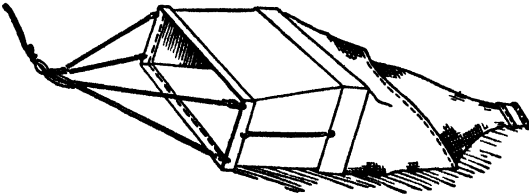


FIG. 16. — Bottom Dredge. After Juday.

The guard is again of canvas while the straining net is of silk gauze. The net is detachable, and net attachments of varying grades of fineness can be attached to the same guard. The net is protected by a

canvas apron that extends from the guard. The bottom opening of the net is reinforced by a band of canvas, and is closed by tying a line about the canvas. The dredge is made to travel along the bottom by attaching a weight to the drag line in front of the mouth of the dredge.

Other types of dredges, as well as traps for plant-dwelling organisms and other special sampling devices, have been described by Birge, Juday, Ekman and others.

Transportation and Preservation of Samples. — Samples of microscopic or larger aquatic organisms can be examined in the field or transported to the laboratory for examination. Many of the aquatic organisms are extremely fragile and decompose quickly when removed from their natural environment. Field examinations are, therefore, recommended, but are commonly impracticable. Microscopic examination must usually be postponed until the samples reach the laboratory.

Transportation of Samples. — Bottle samples that are to be carried to the laboratory are best inserted in canvas bags made to fit the type of collecting bottle adopted, and provided with a cloth handle. If they are to be shipped by express, they should be packed in covered boxes that have compartments lined with suitable packing to prevent breakage. The stoppers should be tied down rigidly by passing a piece of cloth or heavy paper over stopper and neck and fastening it securely around the neck. In cold weather it is necessary to use a lining of felt or other insulating material in the box to prevent freezing. Con-

centrated samples can be placed in small bottles or phials with screw tops or with stoppers tied down as above.

Preservation of Samples. — It is not customary to treat bottle samples with preservative chemicals. When necessary, 5 cc. of chloroform per liter of water will kill the organisms and prevent changes in the microscopic life of the sample between the time of collection and the time of examination. Samples concentrated in the field are naturally more subject to decomposition. They can be preserved by adding a sufficient amount of 95 per cent alcohol to produce a concentration of 75 to 80 per cent in the sample, or by adding a solution of formalin (40 per cent formaldehyde) to give a concentration of 5 to 10 per cent. Preservation is not complete. Many of the organisms, particularly the protozoa, are destroyed during transportation, but some of the carcasses can be recognized even after partial destruction.

Mud samples can be preserved by the addition of an equal volume of strong alcohol. Collections of bottom organisms from which the mud has been removed are commonly treated with formalin to give a concentration of 10 per cent.

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CHAPTER V

EXAMINATION OF SAMPLES

After samples for microscopic analysis have been collected, the nature and abundance of life in the material obtained may be studied in the field or in the laboratory. The methods of examination available are the same in either case. The samples may be examined qualitatively to determine the kind and variety of organisms present, or quantitatively to find the bulk of the catch. In sanitary water analysis, a study is made of the number or bulk as well as of the species of organisms present, because information of both kinds is required in gaging water quality.

Samples that contain luxuriant growths of organisms can be examined directly, whereas those in which there are relatively few organisms must first have the organisms concentrated. Some of the sampling methods, as we have seen, produce a concentrated sample which needs no further treatment unless very high concentration is desired.

THE SEDGWICK-RAFTER METHOD

The method of concentration and examination most frequently used in sanitary water analysis is the Sedgwick-Rafter method. This is prescribed in "Standard Methods of Water Analysis" (1925) and consists briefly of the following processes: the filtration of a measured quantity of the sample through a layer of sand which retains the organisms; the separation of the organisms from the sand by washing it with a small measured quantity of filtered, or distilled, water and decanting; the microscopical examination of a portion of the decanted fluid; the enumeration of the organisms found therein; and the calculation of the number of organisms in the sample of water examined. The essential items of equipment are a filter, decantation tubes, a cell, and a microscope with an ocular micrometer.

Filtration. — Filtration can be accomplished in an ordinary glass funnel 7 or 8 inches in diameter in which the sand is supported upon a plug of rolled wire gauze, or glass wool, but a cylindrical funnel such as shown in Fig. 17 is preferable. The upper part of this funnel, 9 inches in length, has a diameter of about 2 inches. The lower part gradually contracts in 3 inches to a diameter of $\frac{1}{2}$ inch which persists for 2 $\frac{1}{2}$ inches. The capacity of the funnel is 500 cc. The filtering sand is supported

on a perforated rubber stopper pressed tightly into the stem of the funnel and capped with a circular or square piece of fine silk bolting cloth (No. 15X is a good grade of cloth). The bolting cloth covers the perforation in the stopper. The diameter of the cloth circles which may be cut with a wad cutter, or the diagonal of the cloth squares should be slightly less than $\frac{1}{2}$ inch. When moist, the cloth readily adheres to the stopper.

The sand placed upon the stopper should have a minimum depth of $\frac{1}{2}$ inch. The quality of the sand is important. Ordinary sand is unsatisfactory unless very thoroughly washed and ignited. Berkshire sand or pure ground quartz is preferable. The whiteness of these materials is a decided advantage. The necessary degree of fineness of the sand depends somewhat upon the character of the water to be filtered. Sand that passes a sieve having 60 meshes to an inch and is retained on a sieve having 120 meshes is found satisfactory for most samples. Such sand is called 60 to 120 sand. When very small organisms are present, finer sand must be used, such as 60 to 140 sand. The sand used for many years by the author has an effective size of 0.16 mm. and a uniformity co-efficient of 1.4,—that is, 10 per cent by weight is finer than 0.16 mm., and the ratio of the 60 per cent size to the 10 per cent size is 1.4.

This sand is prepared by mixing the following percentages by weight of sieved sand.

TABLE 12 .
COMPOSITION OF FILTER SAND

Meshes per inch	Per Cent of Sample (by weight)
40-60	20
60-80	30
80-100	40
100-140	10

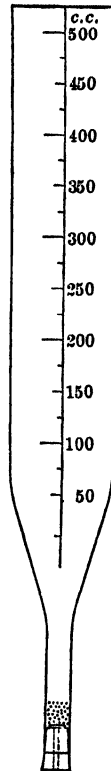


FIG. 17. — Cylindrical Funnel Used in Sedgwick-Rafter Method. (The graduations are commonly omitted except on funnels that are to be used in the field.)

The filter funnel is supported on a ring stand. If many funnels are to be used at the same time they can be conveniently arrayed against

the laboratory wall as shown in Fig. 18, or on a revolving circular frame as in Fig. 19. The filtered water is collected in jars or in a sloping trough draining into a sink. A hinged cover above the filters will exclude dust from the samples.

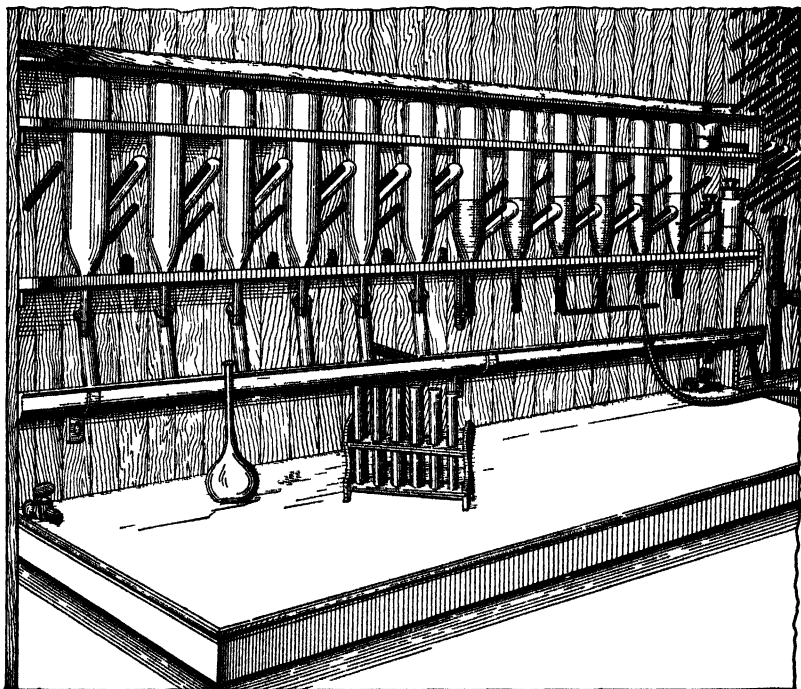


FIG. 18. — Battery of Filters. Sedgwick-Rafter Method.

The sample to be filtered is measured in a graduated cylinder or flask or in the filter funnel itself when it is suitably graduated. The graduated funnel is especially useful for field work, as it saves carrying additional equipment. The quantity of water that should be filtered depends upon the number of organisms and the amount of amorphous matter present. Inspection of the sample enables one to judge the proper amount. Ordinarily 1000 cc. are found satisfactory for ground water and 500 cc. for surface water. In some cases 250 cc. or as little as 100 cc. of surface water are sufficient quantities. When the water is poured into the funnel, care should be taken not to disturb the sand, otherwise organisms may be forced through the filter. It is best to compact the sand by pouring in enough distilled water to fill the neck of the funnel and to add the measured sample before the sand has become uncovered. The accumulation of air in the sand is prevented by

pouring about 5 cc. of distilled water into the funnel and then adding the sand. The funnel should be tilted from side to side to permit the escape of air from the sand. Gravity filtration ordinarily takes place

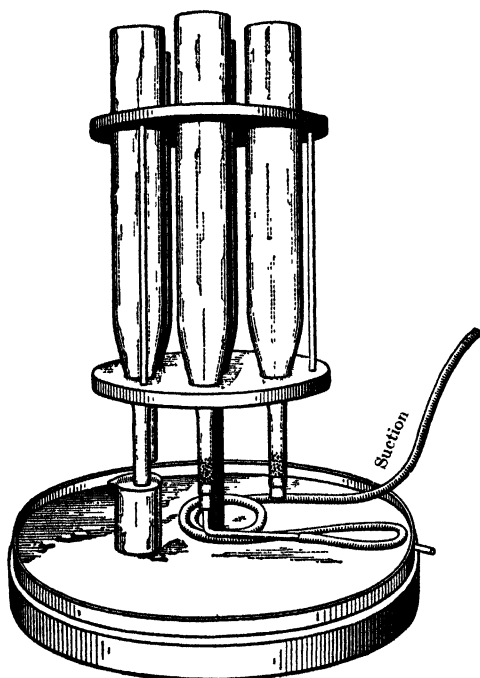


FIG. 19. — Revolving Stand for Filter Funnels. *After Bunker.*

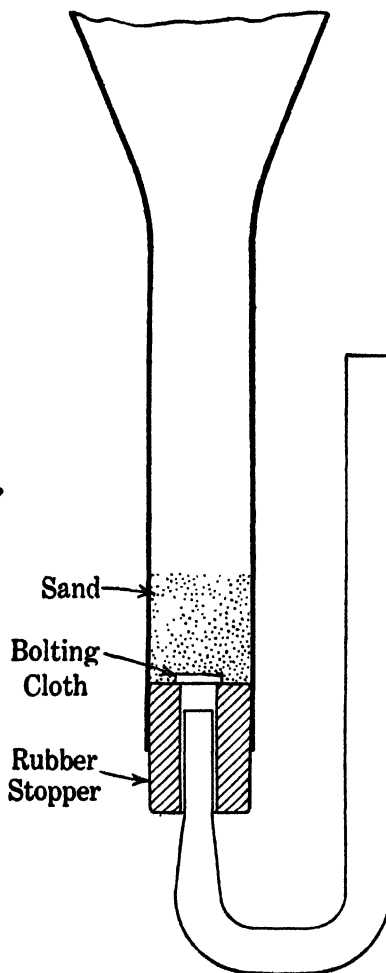


FIG. 20. — Concentrating Attachment.

in about half an hour. During this period the sides of the funnel should be washed down from time to time with a stream of distilled water from a wash bottle. A useful aid in controlling gravity filtration is shown in Fig. 20. With this attachment, filtration cannot proceed beyond the

outlet level of the U-tube. After this level has been reached, the attachment is removed and the filter permitted to drain.

Occasionally a sample is so rich in organisms and amorphous matter that the filter becomes clogged and filters very slowly if at all. It then becomes necessary to scrape lightly the sand surface with a glass rod or, better, to apply suction to hasten filtration. The use of an aspirator not only effects a saving in time but reduces the error caused by the settling of organisms on the sloping surface of the funnel. In order to prevent too violent contact of the organisms with the sand surface and to keep them from being sucked into the sand and destroyed, suction should be discontinued before the water level reaches the constricted section of the funnel. Dr. Frank E. Hale has devised a very convenient arrangement of apparatus for filtration by suction. The filter funnel is pressed into a Bailey crucible holder inserted in the mouth of a suction flask. Filtration is readily controlled by regulating suction, which should not be too strong, since otherwise some of the organisms are lost.

When the sample of water is very turbid the suspended clay will clog the filter so quickly and completely that even suction becomes ineffective. In dealing with water of this type it becomes necessary to prepare the sample for concentration by shaking the sample, allowing the heavy suspended matter to settle, and decanting the supernatant liquid which will contain the bulk of the organisms. The silt is then shaken up with distilled water and allowed to settle, and the clear water is once more decanted. The decanted portions are measured, and filtered after mixing, or they are filtered separately. Some organisms are doubtless caught in the silt and, under certain circumstances, a third elutriation may be required.

Concentration. — Filtration concentrates on the sand the organisms and any other suspended matter contained in the sample. As soon as the sand has drained, the rubber stopper is removed and the sand is washed into a beaker or wide test tube by a measured quantity of filtered or distilled water delivered from a pipette. The amount of water used for washing depends upon the number of organisms collected on the sand. If 500 cc. of the sample are filtered, 5 cc. of wash water are commonly used, thus concentrating the organisms one hundred times. The amount of water filtered divided by the amount of wash water is called the "degree of concentration." This may be varied from 10 to 500 according to the number of organisms in the sample. It is ordinarily 50 or 100.

The container into which the sand and organisms have been washed is shaken to detach the organisms from the sand grains. This is followed by rapid decantation into a second container. Most of the organisms,

since they are lighter than the sand, pass over with the decanted water, while the sand is left upon the walls of the first container. To insure accuracy the sand should be washed a second time and the two decanted portions mixed. If, for example, it is desired to concentrate a sample from 500 cc. to 10 cc., the sand should be washed twice with 5 cc. and the two portions poured together. This will give more accurate results than a single washing with 10 cc.

Instead of washing the sand with a known volume of water, the volume of decanted concentrate may be measured in a graduated pipette fitted with a large bulb. If the amount is not a round number, sufficient distilled water can then be added to bring up the amount to some multiple of 5. If the catch is to be preserved for future examination the preservative should be added prior to measurement.

Examination. For microscopic examination a measured portion of the concentrated fluid is placed in a specially constructed counting cell.

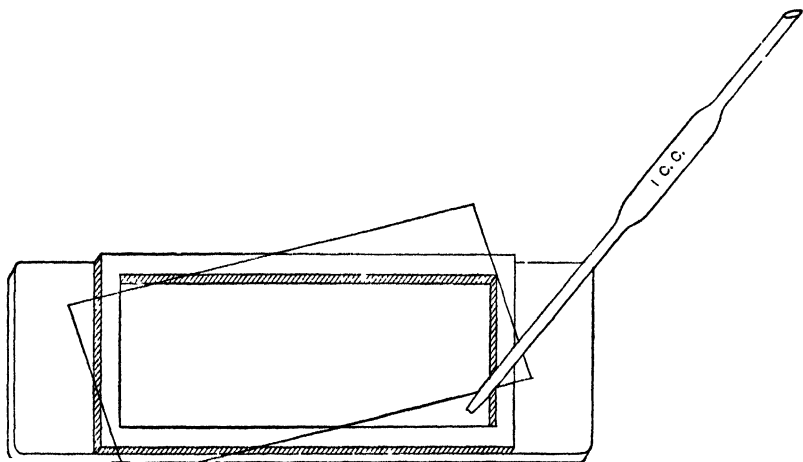


FIG. 21. — Counting Cell, Showing Method of Filling.

This consists of a brass rim cemented with Canada balsam to an ordinary glass slip. The original Sedgwick-Rafter cell is rectangular. Its internal dimensions are: length, 50 mm.; width, 20 mm.; and depth, 1 mm. It has, therefore, an area of 1000 sq. mm. and a capacity of 1 cc. A thick coverglass (No. 3) having dimensions equal to those of the outside of the brass rim (55 mm. by 25 mm.), prevents vibration of the liquid in the cell. Before the cell is filled, the concentrated organisms in the decantation tube are distributed uniformly through the fluid by blowing into it through a pipette. The cell is then filled in such a manner as to distribute the organisms evenly over the entire area. This

is done by placing the coverglass diagonally over the cell so that an opening is left at either end, allowing the water to flow in at one end while the air escapes at the other (see Fig. 21). When the cell is full the coverslip automatically slides into place.

It is not necessary to use a rectangular cell. A circular cell is equally satisfactory, much cheaper, and more easily cleaned. The capacity of the cell is immaterial as long as it is known. A capacity of about 1 cc. is convenient. The depth of the cell must be exactly established. A depth of 1 mm. is commonly used. Baylis uses a cell that holds 10 cc. This permits surveying a larger volume of concentrate for the rarer forms of life. Baylis also omits the coverslip in order to permit shifting from a 16-mm. objective to a 4-mm. objective. This gives the analyst an opportunity to identify those organisms with which he is less familiar and to determine the species as well as the genus of the organisms present. Birge and Juday have used a long, narrow counting cell the width of which is such that it presents one field to view under their binocular microscope. The dimensions of the cell are 63 mm. \times 8 mm. \times 2 mm. deep. The organisms are counted by passing the cell through the field from end to end.

The cell, filled with the concentrate, is placed upon the stage of a microscope and subjected to examination. In order to obtain quanti-

tative estimates of the number or bulk and varieties of organisms present in each field examined, the microscope must be equipped with an ocular micrometer that defines the area of the field. The Whipple micrometer ordinarily used in the Sedgwick-Rafter method consists of a square ruled upon a thin glass disk which is placed upon the diaphragm of the ocular. The square is so dimensioned that with a certain combination of objective, ocular and tube length of the microscope, the area on the stage covered by

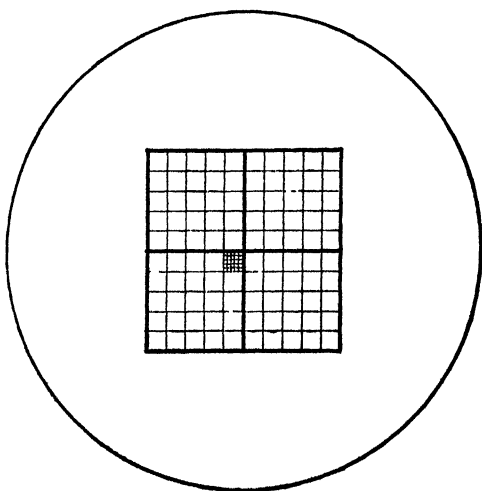


FIG. 22. — Whipple Micrometer.

the ocular micrometer is exactly one square millimeter. Hence, with a cell one millimeter deep, the volume within the outlines of the ruled square is one cubic millimeter. For convenience in determining the

size of the organisms found, the cell is further subdivided as shown in Fig. 22. The use of the Whipple micrometer is discussed in later sections of this chapter and in Chapter VI. The best micrometers are made by engraving, but a serviceable micrometer for occasional use may be made by photography. The square ruled for the Whipple micrometer is 7 mm. on a side.

Enumeration. — Enumeration of the number or bulk and variety of organisms present in the counting cell proceeds in two steps known as *the survey* and *the total count*. The survey serves the purpose of enumerating rarer plankton and those large organisms, such as crustacea, rotifera, and small worms, that move about in the cell or are better seen under very low power. The whole cell should be examined. The results obtained then pertain to the entire volume of concentrate. Bearing this in mind, one can calculate, as explained below, the number or units of "survey" organisms per cubic centimeter of water.

In the total count the organisms or units included within the area of the ruled square are counted. This gives the number or units in one cubic millimeter of the concentrate. The cell is then moved so that another portion of the cell comes into the field of vision and another square is counted. This is continued until a sufficient number of representative millimeter cubes has been examined. It is obviously impracticable to count all of the squares composing the area of the cell; it usually suffices to count ten or twenty squares. In counting, it should be remembered that the cell is one millimeter deep and that some of the organisms are heavy and sink to the bottom, while others are light and rise to the top. The observer should make a practice of changing the focus of the microscope to examine both the upper and lower strata of each cube.

From the number or units of organisms found in the survey and in the total count, it is a simple matter to calculate the number or units present in one cubic centimeter of the original uncondensed sample.

- Let v = the volume, in cubic centimeters, of the counting cell.
 s = the number or units of organisms found in the "survey."
 n = the number of squares counted, i.e., the number of cubic millimeters of the concentrate actually examined.
 t = the total number or units of organisms found in all of the squares counted in the "total count."
 f = the volume, in cubic centimeters, of the sample filtered or concentrated.
 w = the volume, in cubic centimeters, of the concentrate, i.e., of the water used in washing the sample.

Then the number or units of organisms per cubic centimeter (N) is given by the equation:

$$N = \frac{w}{fv} s + \frac{1000 wt}{fn} = \frac{w}{f} \left(\frac{s}{v} + 1000 \frac{t}{n} \right).$$

If, for example, 250 cc. of water were filtered and 15 cc. of water were used for washing the sample; if, furthermore, the cell held 1 cc. and 10 squares were counted:

$$N = \frac{15}{250} \left(\frac{s}{1} + 1000 \frac{t}{10} \right) = 0.06 s + 6 t.$$

The coefficients obtained for s and t are known as the "multipliers." The methods of reporting the results of enumeration are discussed more fully in the next chapter.

Sources of Error. — The operations of the Sedgwick-Rafter method involve several sources of error. They may be classified as follows:

1. Errors in sampling.
2. Funnel error, or the error caused by the organisms adhering to the sides of the funnel.
3. Sand error, or the error caused by imperfect filtration.
4. Error of disintegration, due to the breaking up of organisms on the surface of the sand.
5. Decantation error, or the error caused by the organisms adhering to the particles of sand, and by the water used in washing the sand being held back by surface tension during the process of decantation.
6. Pipetting error, or the error due to sampling the concentrate.
7. Errors caused by the organisms not being distributed uniformly through the cell.

Errors in Sampling. — These errors arise chiefly from the fact that organisms vary in specific gravity and in their behavior toward light. If the bottle containing the sample is allowed to stand even for a short time, some of the organisms sink to the bottom, some rise to the surface; some collect on the side of the bottle toward the light, others shun the light as much as possible; some attach themselves quite firmly to the sides of the glass. Evidently, the bottle must be shaken before the portion for examination is withdrawn. Errors in sampling are common but, to a great extent, avoidable.

Funnel Error. — The funnel error, which is due to the organisms settling upon and adhering to the sloping sides of the funnel, varies with the character of the water filtered. It is highest in the case of samples rich in cyanophyceæ and amorphous matter. These, being of a somewhat gelatinous nature, adhere readily to the glass and produce a rough

and sticky surface upon which other organisms lodge. If the funnel is wet when the sand is put in, some of the sand grains are likely to adhere to the sloping walls and cause an increase in the deposition of organisms. The funnel error is less in cylindrical funnels than in ordinary flaring funnels. Slow filtration, whether due to the shape of the funnel or to the characteristics of the sample filtered, increases the error — indeed, it may be said that the funnel error is almost proportional to the time of filtration. Numerically, the funnel error varies from 0 to 15 per cent. A long series of experiments with waters that differed greatly in character gave an average funnel error of 1 per cent for organisms and 3 per cent for amorphous matter. The funnel error can be reduced by increasing the rate of filtration through the use of an aspirator, by washing down the sides of the funnel with distilled water during filtration, and by cleaning the funnel scrupulously before use.

Sand Error. — The sand error, which results from imperfect filtration, depends upon the character of the organisms, the size of the sand grains, and the depth of the sand. In selecting the sand a balance must be struck between two contrary requirements. The sand must be fine enough to be an efficient filtering medium and yet coarse enough to settle readily in the decantation tubes. A $\frac{1}{2}$ -inch layer of the sand described in Table 12 does not give a sand error greater than 5 per cent unless the water contains exceptionally small organisms. When these are present in large numbers, the error from incomplete filtration may be as great as 50 per cent. The effect of the size of the sand grains on the sand error is well illustrated by the following table, compiled from experiments by Calkins on the filtration of water containing yeast cells and starch grains:

TABLE 13
SAND ERROR OBTAINED IN FILTERING YEAST CELLS AND STARCH GRAINS

Size of Sand	Percentage Sand Error	
	Yeast Cells	Starch Grains
40-60	21.6	4.4
60-80	8.7	7.3
80-100	5.3	7.4
100-120	3.3	1.2

Most of the organisms that pass through the sand do so during the early part of filtration, that is, before the sand has become compacted

and before a surface mat of organisms is formed. The sand error is reduced considerably if, before the sample is poured into the funnel, the sand is compacted by passing through it some distilled water, using the aspirator to increase compacting.

Errors of Disintegration. — Many of the microscopic organisms are extremely delicate. They are very susceptible to changes in temperature, pressure, and light. As soon as a sample of water has been collected in a bottle, some of the organisms begin to disintegrate; and if the sample stands long before examination or is subjected to the joltings of a long trip, some of the organisms break up and become unrecognizable. The process of filtration helps to disintegrate them by bringing them into violent contact with the surface of the sand, but the method of concentrating the sample in which filtration is arrested as described above reduces this error to some extent. The errors due to disintegration during transit and before examination can be avoided only by making the examination in the field. This is often necessary, particularly when one is searching for such delicate organisms as *Uroglenopsis*. The errors of disintegration during filtration cannot be entirely avoided; but if examination of the concentrate is supplemented by direct examination of the water, gross mistakes are prevented. After a little practice, *Uroglenopsis*, *Dinobryon*, and other forms are detected in the sample with the naked eye. They can be taken up with a pipette and transferred to the stage of the microscope for more definite identification. This direct examination is important and should always be made, but its value is qualitative and not quantitative.

Decantation Error. — The decantation error depends to a great extent upon care in manipulation. When one attempts to separate the organisms from the sand by agitating with distilled water in one test tube and decanting into a second tube, some of the organisms remain behind, attached to the sand grains, and, what is quite as important, some of the water used in washing remains behind.

The two errors are compensating. If the sand retains a larger percentage of organisms than of water, the result is too low; if it retains a larger percentage of water than of organisms, concentration becomes too great and the result is too high. With the fractional method of washing the sand and with due care in manipulation, the decantation error should not exceed 5 per cent.

Errors in Pipetting. — Only a portion of the concentrate is actually placed in the counting cell. This involves an error in sampling which is reduced to a minimum by blowing air into the concentrate through the pipette before withdrawing the sample. Allen has investigated the pipetting error in combination with the errors of the cell discussed below.

An analysis of his data shows that the probable error of pipetting and cell is about 10 per cent.

Errors in the Cell. — The errors due to the unequal distribution of the organisms over the area of the cell are extremely variable and cannot be readily stated in figures. If the concentrate is evenly mixed and well distributed over the cell, if the count is made as soon as the material in the cell has settled, and if a large number of squares is counted, the error is reduced to a minimum. If a sample happens to contain such motile organisms as *Trachelomonas* or *Euglena*, they often collect at the edges of the cell in search of air; or, if the cell stands in front of a window for any length of time, organisms sensitive to light migrate from one side of the cell to the other.

Precision of the Sedgwick-Rafter Method. — Examination of hundreds of samples has shown that results obtained by the Sedgwick-Rafter method are usually *precise* within 10 per cent, i. e., two examinations of the same sample seldom differ by more than this amount. Whereas the precision is relatively high, the accuracy (exact conformity of the test findings to actual conditions) varies greatly with the character of the organisms in the water examined. On account of unavoidable errors, care should be taken not to imply fictitious accuracy in tabulating the final results. No decimals or fractions should be used.

OTHER METHODS OF EXAMINATION

The complete Sedgwick-Rafter method was developed particularly for the microscopic analysis of bottle samples. It is more especially a laboratory method although it can be employed in the field. The concentration equipment used is somewhat fragile. Adaptations of it for field conditions have, therefore, been described in the preceding chapter. Concentration may be accomplished also by means other than sand filtration. The plankton-net method has already been mentioned, and other ways of condensing the catch are described below.

The principles of examination and enumeration of the Sedgwick-Rafter method are in common use whenever it is purposed to obtain complete information about the number, or units, and varieties of organisms present in the sample examined. Sometimes all of this information is not required or the means for obtaining it is too laborious. Under these circumstances quantitative estimates of the bulk of the plankton catch can be made by direct estimate or gravimetric methods which will be described below. Certain modifications of the Sedgwick-Rafter method are also included. When bottom sediments, rather than water samples, are to be examined, special methods of examination must be resorted to.

The Kofoid Method. — Some observers prefer using, in place of the Sedgwick-Rafter funnel with its sand column, an ordinary laboratory funnel with hard-surface filter paper, such as No. 575 Schleicher and Schüll or No. 50 Whatman. The sample is filtered as in ordinary analytical work until about 5 cc. remain in the funnel. This portion of water, containing a large percentage of the organisms, is pipetted out or poured into a tube, and the filter paper is then washed by directing a stream of distilled water against the sides. Ten cubic centimeters are usually required to dislodge the organisms from the paper. The resulting concentrate is added to that obtained previously. After the volume of the catch has been measured it is examined according to the Sedgwick-Rafter method. The filter-paper method was first used by Kofoid in his studies of the plankton of the Illinois River. Results comparable with those of sand filtration are obtained.

In order to expose the entire surface of the filter paper to the jet of wash water, the paper may be folded as a double cone. This is done as follows: Fold the paper once across; open out; pinch crease at the periphery and break crease in center; bring ends together so that they overlap and form a double cone; insert in funnel.

The Plankton-Net Method. — The plankton-net method for the collection of microscopic organisms yields samples that require no further concentration. The organisms obtained, however, it must be repeated, constitute only the larger varieties of microscopic life; the smaller ones escape through the bolting cloth. This is a basic limitation of the method. Comparisons of the net method with the Sedgwick-Rafter or Kofoid method cannot be drawn except on the basis of the larger plankton collected.

Identification and enumeration of the concentrate obtained by the net method proceeds in the same way as for the Sedgwick-Rafter method unless only a rough measure of the bulk of the catch is desired. The latter is obtained by allowing the concentrate to settle for 24 or 48 hours and then measuring the volume of sediment collecting. With turbid waters the amount of silt in the concentrate must be estimated and subtracted. The catch is commonly expressed in cubic centimeters of plankton per cubic meter of water filtered, i.e., parts per million by volume. Settling can be hastened by centrifuging; at the same time, a more compact sediment is obtained. Since different concentrating methods will yield different results, the method of concentrating should always be specified.

Centrifuging Methods. — The centrifuge has been used in various ways to concentrate microscopic organisms for examination. Its use for this purpose has not yet been standardized and different observers

have employed machines varying in design and operation. Speeds of 1000 to 50,000 revolutions have been used and the time of run has varied from one to many minutes. If comparable results are to be obtained from different machines, both the centrifugal pressure exerted and the time of concentration must be regulated. The pressure obtained by centrifuging is given by the formula:

$$p = s \frac{\pi n}{60} (r^2 - r_0^2),$$

where p = pressure in dynes per square centimeter.

s = specific gravity of the tube contents

n = number of revolutions per minute.

r = distance in centimeters from axis of rotation to bottom of tube.

r_0 = distance in centimeters from axis of rotation to top of liquid.

A simple laboratory centrifuge holding 20 cc. in each tube is shown in Fig 23

Use of Centrifuge for Direct Estimation of Volume of Catch — The most general application of the centrifuge is in connection with the plankton-net method for the further condensation of the catch and direct estimate of the volume obtained. When gravity alone is relied upon to condense the catch, the results obtained vary greatly with the character of the plankton and silt. This is shown by the reduction in volume observed when different concentrates are centrifuged. In general, filamentous algæ and diatoms and flocculent débris show greater shrinkage than killed protozoa, rotifera, and heavy silt. The decrease in volume varies from a few per cent to over 70 per cent. Centrifuging, therefore, gives a better estimate of the true plankton volume. Results are expressed in cc. per m.³ and must be corrected for silt. In his studies of the Illinois River, Kofoid used centrifugal pressures of 1.4 million dynes per square centimeter for one minute.

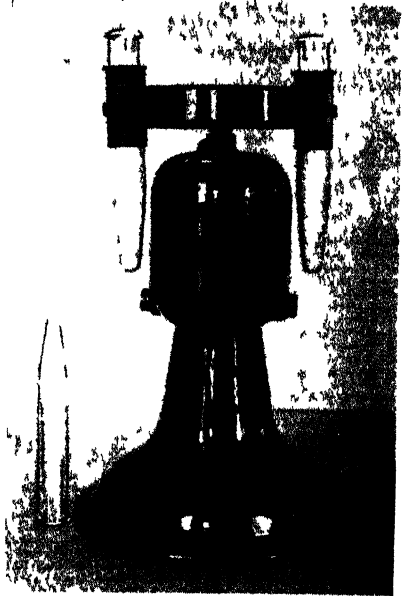


FIG. 23 — Laboratory Centrifuge

Ward employed higher values for longer intervals of time on Lake Michigan, and so did Juday on Turkey Lake.

Use of Centrifuge in Connection with Counting Methods. — The centrifuge has also been used to concentrate organisms directly from samples of water, the condensed material being subjected to microscopic examination and enumeration.

In their studies of the nannoplankton in the Finger Lakes of New York, Birge and Juday employed a common type of centrifuge carrying 15-cc. tubes. The machine rotated at a rate of 3600 R.P.M. Sedimentation was completed usually in 6 minutes, although a second and third run were often necessary. The settled material was removed for examination by means of a pipette. Results were expressed in c.c. per m.³.

An objection to the use of the ordinary centrifuge is the fact that those organisms whose specific gravity is less than one rise to the top of the water, where they are not readily gathered by pipetting. Reduction of the density of the liquid by preservatives such as alcohol is sometimes advisable.

Houston's Use of the Centrifuge. — Instead of using counting methods, Houston in some of his investigations has prepared photomicrographic

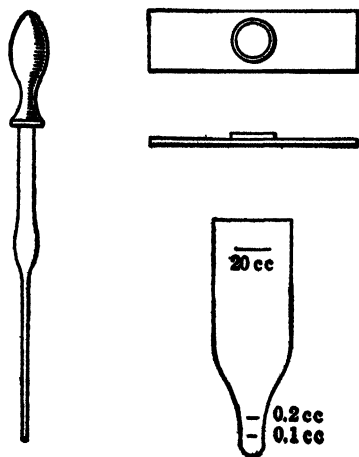


FIG. 24. — Pipette, Slide and Tube Used in Obtaining Plankton Concentrates for Quantitative Photomicrography. After Houston.

records of the suspended matter in the London waters. Photographs are taken of samples concentrated by centrifugal methods. Part of Houston's equipment is illustrated in Fig. 24. His technique may be summarized as follows: Place 20 cc. of the sample in the tube; centrifuge until all suspended matter is driven to bottom; remove all water above 0.2-cc. mark; mix deposit and, by means of the pipette, transfer 0.1 cc. of the suspended matter to the photographic cell; add a trace of formalin, and photograph, using a magnification of 50 diameters. About 1/20 part of the suspension falls in the photographic field. This corresponds to the suspended matter in $\frac{1}{2}$ cc. of the original water.

Birge and Juday's Use of the Centrifuge. — The Wisconsin Geological and Natural History Survey has made an extensive study of the quantity of nannoplankton contained in water by determining its dry weight

per unit volume of water. Since the weight of the individual organism is extremely small, it was necessary to concentrate the organisms suspended in 1000 to 1500 liters of water. A De Laval centrifuge and filter was used for this purpose. Tests showed that 98 per cent of the algae and protozoa lost by the plankton net were recovered by the centrifuge. Twenty-five to fifty per cent of the bacteria were also removed. In order to extend this use of the centrifuge to field work, Foerst constructed two smaller units operated at 20,000 and 32,000 revolutions per minute, respectively. Juday has used these machines with great success to concentrate plankton for enumeration and chemical analysis. The construction of the smaller machine is shown in Fig. 25. The principle of operation is the same as that of a "milk clarifier." The organisms are thrown on to the inclined wall of the revolving bowl whence they are removed for bulk measurement, chemical analysis, or enumeration and identification. The clarified water escapes over the rim of the bowl. Normal quantities of water are sufficient for purposes of enumeration.

Examination of Bottom Sediments. — The study of organisms found in sludge and other bottom sediments requires a technique very different from that used in the examination of the supernatant water. The method developed by the United States Public Health Service in its investigation of the Ohio River is given by Purdy (Public Health Bulletin 131) as follows:

"The laboratory examination of bottom sediments consists in noting the color, the odor, and the consistency or 'streak.' Following this, the mud is carefully washed through a fine-mesh sieve. This is accomplished by placing the mud in some such vessel as a 'moist chamber' — which has perpendicular sides — running tap water into it and agitating

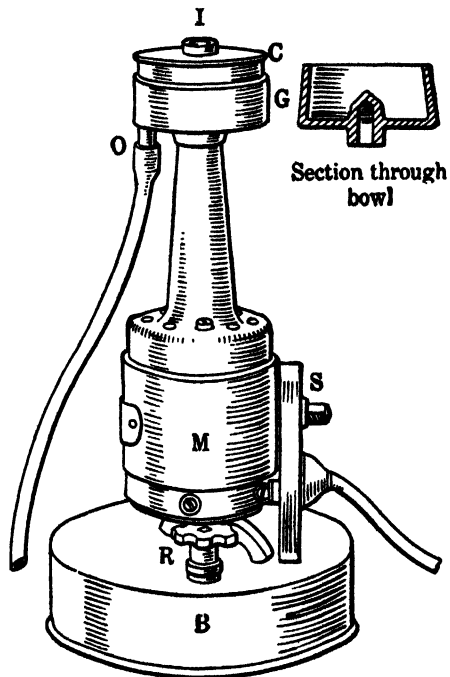


FIG. 25. — Foerst Centrifuge. After Juday.

B, base; *M*, motor; *R*, rheostat wheel; *O*, outlet tube; *G*, guard; *C*, cover; *I*, inlet tube; *S*, switchboard.

and rotating the vessel until the water is very turbid with mud in suspension. This is then poured through the sieve. More tap water is then added and the process repeated as often as necessary until all the mud has been passed through the sieve, whose fine meshes retain the larger organisms present (worms, insect larvæ, mollusks, etc.) and the various kinds of disintegrating organic material, chiefly fragments of leaves, plants, seeds, small sticks, molts of water animals, etc. This is essentially a process of filtration and the 'catch' in the sieve is transferred to a vessel of clean water for convenient study. Results are tabulated under appropriate headings. No quantitative estimates are attempted aside from using a sample of constant size (200 cc.). Organisms present are merely counted. Disintegrating organic matter is recorded as to predominating kinds and relative abundance."

In practice, the "sieving" of the organisms is best accomplished in the field and the catch is preserved in water with 10 per cent formalin. A sieve with 30 meshes to the inch is used.

TECHNIQUE OF THE MICROSCOPE

The selection of a microscope for the study and enumeration of microscopic organisms will be dictated by the resources at the command of the observer and by the importance attached to the examinations. Of all the instruments used in the pursuit of scientific studies, none has commanded in its development deeper study or a higher degree of mechanical skill than the compound microscope. Accordingly, it is obtainable in a variety of models which possess varying degrees of optical and mechanical perfection. The best instruments are the ones that naturally lend themselves to greatest satisfaction in use and that produce results with the least strain upon the operator. While an expensive equipment is not necessary for the numerical estimation of the common microscopic organisms found in water, it is poor economy to do without those adjustments which measurably add to convenience and conserve time.

Necessary Equipment. — Among the various makes and models of microscopes there are certain parts of the stand that are always supplied and are essentially the same, for they serve fundamental needs. Such are the base, pillar, stage, arm, mirror, and focusing adjustments. There are other parts that vary with the models, concerning which a choice must be made. These will be discussed in succeeding paragraphs. Figure 26 presents the parts of an instrument properly equipped for routine study of microscopic organisms.

Draw Tube. — The draw tube is a telescopic extension of the body tube and is calibrated in millimeters to represent the mechanical tube length between the shoulder of the eyepiece and that of the objective, including the length of the revolving nosepiece. Microscopes with fixed tube length are not desirable for the reason that it is frequently neces-

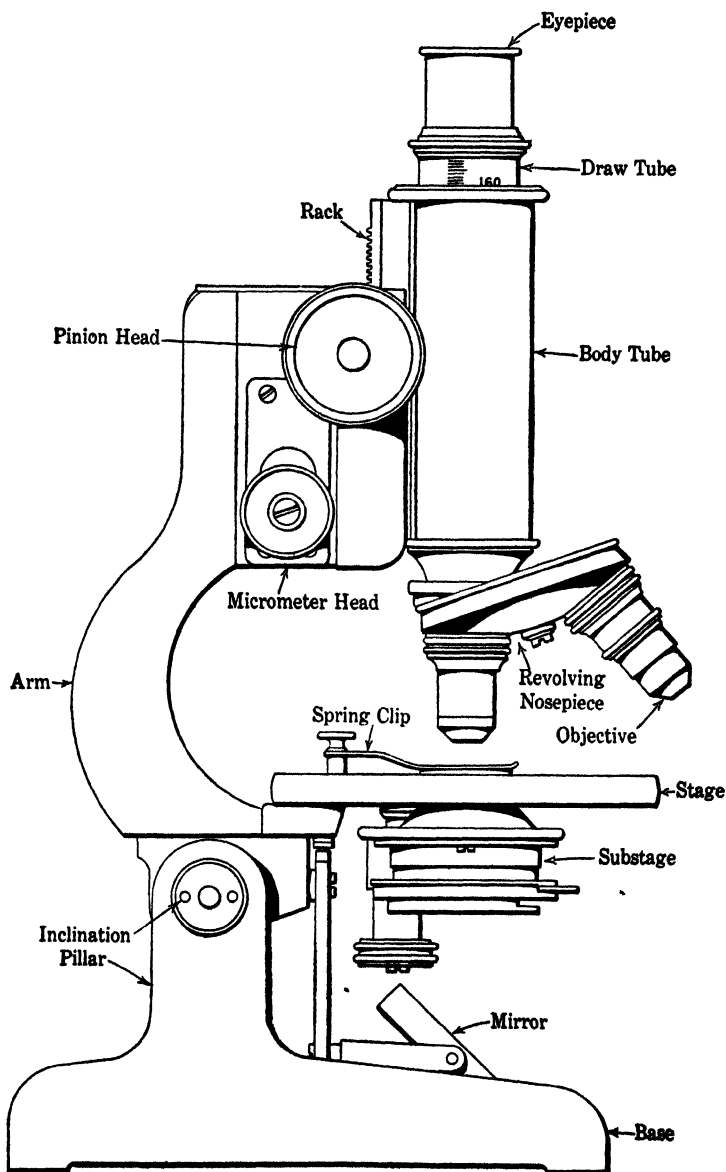


FIG. 26. — Compound Microscope Suitable for the Examination of Water.

Courtesy of Bausch and Lomb

sary to modify the magnification slightly by means of the draw tube in order to obtain a desired value for the ocular micrometer.

Two standard tube lengths exist, the short 160 mm. and the long 216 mm. The former is the more usual for it lends itself to the construction of a shorter, more compact stand.

Lenses. — The revolving nosepiece, while not a necessity, is a great convenience inasmuch as it keeps ready for immediate use two objective lenses. Both of these are needed. The lower-power objective should be a 16-mm. lens, with which enumeration of forms is made. The higher should be a 4-mm. lens which is often employed to study structure and detail and assist in identification of species.

The rating of the lenses is stated either in terms of the equivalent focus of the lens system or in terms of its magnifying power. Thus with a draw-tube length of 160 mm. common ratings are $10\times$ or 16 mm. and $43\times$ or 4 mm. It is not necessary to employ lenses having the highest degree of correction for color aberration. The usual achromatic lens sold by reputable makers is sufficiently well corrected, does not require a compensating eyepiece, and possesses sufficient flatness of field.

The eyepiece, or ocular system of lenses, is a simpler combination than the objective, but should be comparable in quality with the latter in order not to impair optical efficiency. The usual type is the Huygenian eyepiece, named for Huygens, who first used it. The lenses are two in number, a collective lens at the lower end, and an eye lens at the upper end. Between the two, and located at the focal point of the eye lens, is a perforated diaphragm which limits the size of the field and upon which rests the ocular micrometer. The construction and function of the eyepiece, as well as other parts of the microscope, are shown diagrammatically in Fig. 27. Eyepieces are usually rated according to the number of times they magnify the image formed by the objective. Thus a $7.5\times$ eyepiece magnifies the image 7.5 times. This is the power best adapted to work with microscopic organisms as it enables a field 1 mm. square to be covered when working with a 16-mm. objective.

Ocular Micrometer. — A necessary part of the equipment is an ocular micrometer by means of which the sizes of organisms may be computed. There are many kinds, but that used in the Sedgwick-Rafter method is the Whipple micrometer (Fig. 22) made in the form of a square ruled into subdivisions. A description of this is given on page 96. It is placed, ruled side down, upon the diaphragm of the eyepiece, i.e., at the focal point of the eye lens, the image of the ocular micrometer thereby appearing superimposed upon that of the objective field.

Objective Micrometer. — The ocular micrometer must be accurately calibrated to establish the value of its rulings. For this purpose an objective, or stage, micrometer should be available. This consists of a thin glass disk mounted permanently upon a glass slide. On the under side of the disk is etched an accurate linear scale, usually 1.0 mm. divided into hundredths. The procedure of calibration is given on page 119.

The Micrometer Head. — The micrometer head which operates the fine adjustment is usually made so that one complete revolution moves

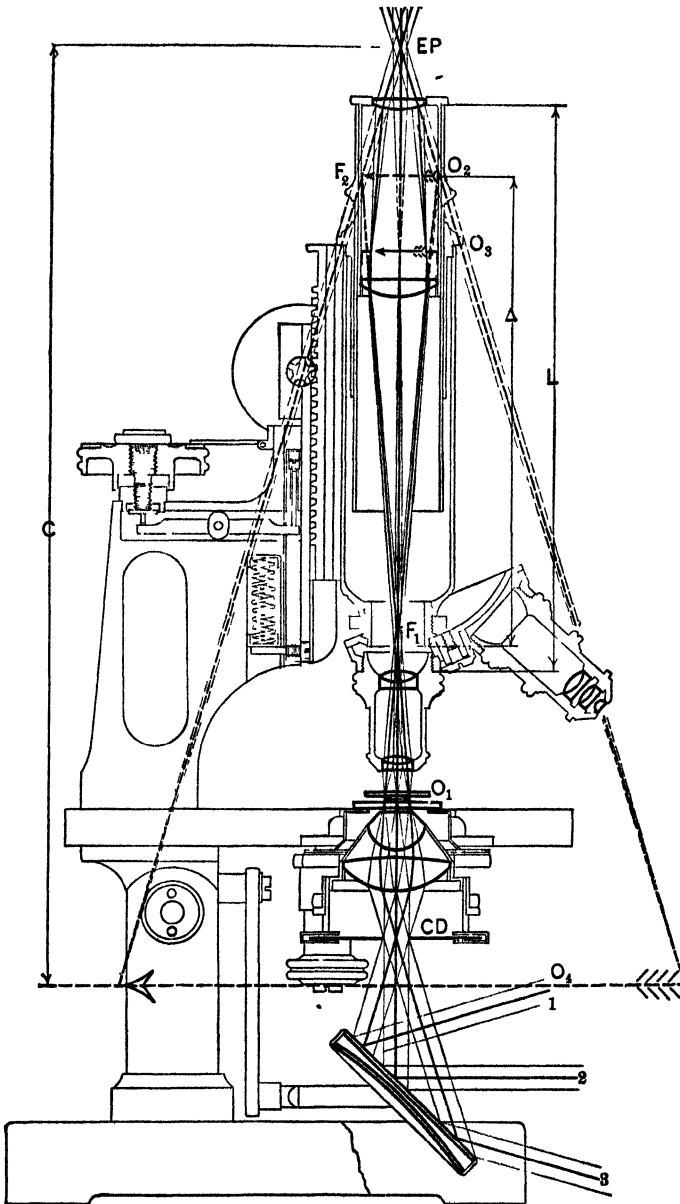


FIG. 27. — Optics of the Compound Microscope. After Bausch.

F_1 , upper focal plane of objective; F_2 , lower focal plane of eyepiece; Δ , optical tube length = distance between F_1 and F_2 ; O_1 , object; O_2 , real image in F_2 , transposed by the collective lens to O_3 , real image in eyepiece diaphragm; O_4 , virtual image formed at the projection distance C , 250 mm. from EP, eye-point; CD, condenser diaphragm; L , mechanical tube length (160 mm.); 1, 2, 3, three pencils of parallel light coming from different points of a distant illuminant, for instance a white cloud, which illuminate three different points of the object.

the body tube up or down a definite distance, generally 0.25 mm. or 0.125 mm. In this way values are obtained for the third, or vertical, dimension of objects. Standard procedure for the Sedgwick-Rafter method calls for estimation of the third dimension of organisms in connection with the determination of cubic standard units. Ordinarily this will be done without actual measurement of the third dimension. The thickness of the organism will be estimated from measured dimensions in the horizontal plane and from a knowledge of the shape of the organism. To obtain the third dimension accurately, it is necessary that the total motion of the micrometer head be subdivided into units. On the more expensive instruments the head carries a graduated revolving drum the divisions of which are submultiples of the distance through which the body tube moves with one revolution of the head. Cheaper instruments have only a rating for the distance covered by one complete revolution. In such a case the drum must be calibrated for intervals of this distance.

The Substage and Condenser. — The substage with its various attachments is a most valuable adjunct of the microscope and a necessary one in all but the simplest examinations. It carries an upper and a lower iris diaphragm for regulating the amount of light, a condenser for regulating illumination, and a carrier for holding either blue glass in connection with the use of artificial light or a dark ground stop. The least expensive form of substage is that which consists of a sleeve attached to the stage, into which the condenser is slipped. Other forms are made with screw, or rack and pinion, mechanism for focusing the condenser and swinging it to one side when not in use.

The purpose of the condenser is to control a most important part of microscopic technique, the illumination of objects. To this end the condenser reduces the volume of light but increases its intensity and produces a cone of light having an angular aperture equal to that of the objective. For the best work, condensers should be chromatically and spherically corrected, but the usual Abbé condenser is neither; yet it gives results much superior to those obtained without any condenser. The simplest type, that with two lenses and a numerical aperture of 1.20, suffices for the needs of the Sedgwick-Rafter method.

Accessory Equipment. — There are many accessories of the microscope, some designed to extend and improve microscopic vision, others to make matters of microscopic technique either more accurate or less laborious. Certain of these accessories are applicable to the study of organisms found in water and will be briefly referred to. They are not necessary for routine examinations but constitute useful equipment for special studies.

Binocular Eyepiece. — Binocular microscopes overcome certain limitations which the monocular type imposes. When both eyes are employed for observation they function in a natural way without convergence, as though looking at more distant objects. As a result, there is freedom from eye strain and general fatigue which come from prolonged periods of monocular vision. The observer can apply himself more closely to examination of objects and will have the benefit of the

stereoscopic effect that throws objects into relief and renders detail more distinct.

Binocular microscopes equipped with all the usual adjustments are expensive, and some of them require the use of paired objectives and eyepieces. The binocular eyepiece, one of the latest improvements of the microscope, can be purchased separately and is easily attached to many of the models, thus making possible the use of the instrument as either a monocular or a binocular microscope. The increase in tube length which results from the use of the binocular eyepiece nearly doubles the magnification.

Demonstration Eyepiece. — A valuable accessory for class work, or for comparative observations by two people at the same time, is the demonstration eyepiece. It is readily attached to any microscope in place of the usual eyepiece. One observer views the field from the customary position through a vertical eyepiece; the other views the field at the same time through a horizontal eyepiece which is carried in the end of a side tube about 6 inches long. There is a pointer mounted on a universal joint in the side of the eyepiece which can be used by either observer to bring to the attention of the other objects or details of structure.

Oil-Immersion Objective. — When a magnification greater than 400 to 500 times is desired, this is best secured by the use of the oil-immersion objective, which produces an image from 90 to 100 times larger than the object. This objective is obtainable with varying degrees of color correction. The achromatic immersion objective is of a sufficiently high order of correction to meet the demands of all but the finest kinds of work. It has an equivalent focus of 1.9 mm. and magnifies 97 times. With the $7.5\times$ Huygenian eyepiece and this objective, the final magnification of the image is 727 times; with the $10\times$ eyepiece it is 970 times; while with the $20\times$ compensating eyepiece it is 1940 times. This is close to the highest limit of magnification, namely, that obtainable with a highly corrected immersion objective and a $25\times$ compensating eyepiece.

The use of a high-power objective calls for certain procedure not demanded with low and medium powers. A drop of cedar immersion oil must be placed between the lens and the coverglass. This eliminates refraction of light rays by the latter, as coverglass and cedar oil have nearly the same refractive index and dispersive qualities, which in turn are close to the qualities of crown glass used in the lenses. Immersion objectives are, therefore, not affected, as are dry objectives, by slight variations in the thickness of coverglass, there being no intervening air space between the object and the lens. Without the use of oil no image can be seen, the light rays in passing from the coverglass to the air being refracted outside the opening of the lens. Immersion oil should, also, fill the space between the slide and the lens of the condenser. It is necessary to adhere closely to the tube length for which the objective was corrected in order to secure optical perfection.

Mechanical Stage. — The mechanical stage is designed to make possible the orientation of the microscopic field, a procedure that is useful for special studies and for checking counts of particular fields. Rack

and pinion movements permit motion of the slide in two directions, and millimeter scales record the exact latitude and longitude. For routine examinations this accessory is not necessary and, in fact, is more likely to impede operations than to promote them. Mechanical stages are furnished as part of the equipment of some microscopes, in which case the movement is inherent in the stage. The attachable form of mechanical stage clamps to the plain stage and can be dispensed with when not desired.

Camera Lucida. — The camera lucida is a valuable accessory for making drawings of objects on an enlarged scale. The image appears to be projected upon paper, where it is traced, while the observer remains at the microscope, the image of the field and of the paper being superimposed by the aid of a mirror.

The Abbé camera lucida is the most satisfactory form. It is compact in construction, is easily attached to the microscope tube, and gives superior results. If unfamiliar with the camera lucida, one should acquaint oneself with its construction and use by referring, before

attempting to operate it, to some standard work on the microscope and its accessories. Such a course will effect economy of time and effort, for considerable skill is required at first in adjusting the apparatus.

Photomicrographic Apparatus. — The most accurate record of microscopic objects is that made by photography. To one possessed of a knowledge of photographic processes, the pursuit of photomicrography is a most interesting and fascinating occupation. One who is not familiar with the principles of photography is likely to have little appreciation of the technical difficulties to be encountered and will meet with failures and disappointments until these principles are mastered.

Elaborate apparatus is not necessary to obtain successful and worthwhile reproductions. Figure 28 shows how a box camera may be fitted over the eyepiece of the microscope. Photographic lenses are not

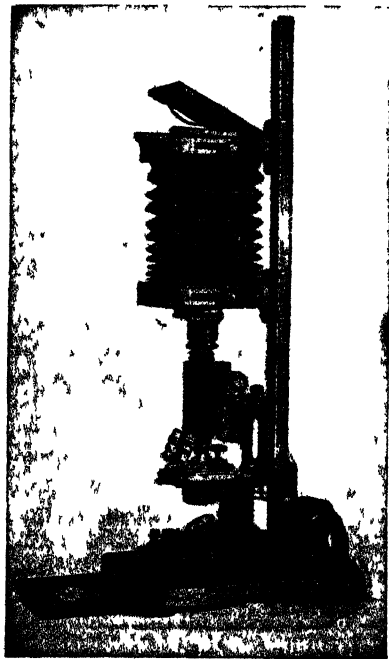


FIG. 28. — Photomicrographic Camera.

required. The principal desiderata are that the whole equipment be rigidly mounted and placed on a base free from vibration, that the optical centers of the camera and microscope be in coincidence, that the camera have a bellows extension and a focusing screen, and that a strong source of artificial illumination be available. Either a hori-

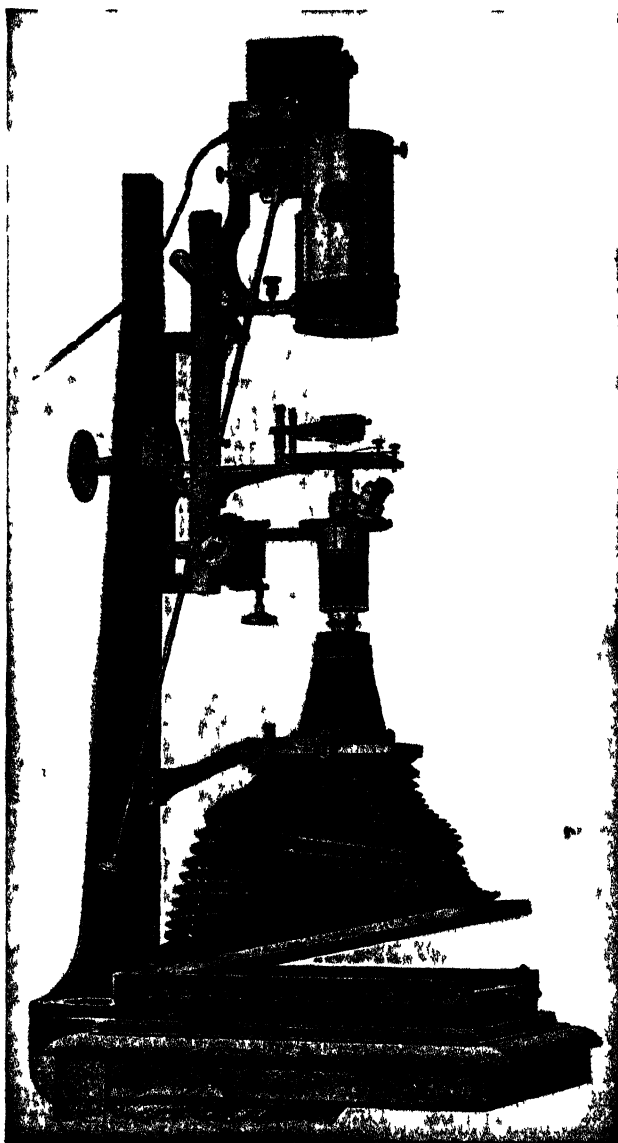


FIG 29 — Edinger Drawing and Projection Apparatus Adapted for Photomicrography.

zontal or a vertical arrangement of the camera may be used, but the vertical arrangement is better where simple apparatus is used and where space is limited. It is also better for photographing the liquid contents of cells, for the latter will be in a horizontal position on the stage.

For obtaining the most satisfactory photomicrographs, particularly with high magnifications, a more flexible outfit is needed than can be supplied by means of a box camera. Adjustments are needed for manipulating magnification, illumination, etc. There can be secured from optical companies models that are built on a special portable stand, and that contain microscope, camera, artificial source of light, and all important adjustments. Such an outfit is that shown in Fig. 29. This instrument can be used for drawing as well as for photography.



FIG. 30. — Bausch and Lomb's Portable Microscope.

Portable Microscopes. — Although most microscopes are supplied with rugged wooden cases that make possible the transportation of the instrument into the field, there are times when the bulk and weight of the apparatus offer a distinct handicap to its use. In such circumstances a more compact outfit is desirable. That shown in Fig. 30 supplies the need. The instrument has a small stage and folding base, and weighs in the case about $8\frac{1}{2}$ pounds. The illustration shows this model equipped with Abbé condenser, three objectives, and two eye-

pieces. Any of these may be omitted from the outfit as purchased, if they are not needed.

Manipulation of the Microscope. — Proper manipulation and care of the microscope can be appreciated only after acquiring familiarity with its parts and its use, and then only by realization on the part of the worker of certain fundamental considerations. Some of these are presented here for the benefit of the beginner in microscopy. For the details of manipulation and the function of different parts of the microscope, he should carefully study the manual which accompanies each instrument and consult standard books on the subject.

In the first place, let the impression be firmly fixed that the microscope is an apparatus of many parts, designed to work together for purposes of precise observations. It must, therefore, be handled with care and when not in use should be protected from harmful influences. The repair or readjustment of damaged parts should be attempted only by one skilled in the making of optical instruments.

Position. — In order to pursue microscopic studies with comfort and a minimum of fatigue, it is necessary that the position of the observer and of the instrument be correct. The seat used should be of such height that the observer sits nearly upright. Strain upon the muscles of the neck and back is relieved by the use of a seat with a back, also by resting the arms upon the bench. The microscope should stand not far from the edge of the bench. Use of the inclination joint to throw the optical axis off the vertical makes for ease of observation, but this practice should be avoided with liquid preparations in a cell; the cell contents are liable to concentrate near the side.

Use of the Eyes. — The distance of the eye from the ocular is determined by the magnification of the latter and is correct when shadows or colors do not appear in the field. The eye-point is closer to the lens with high-power oculars. Which eye to use will often be determined by their relative strength, but the use of the left leaves the right free for use in making sketches or notes without turning the head. In order to relieve strain and muscular fatigue, the habit of keeping both eyes open should be cultivated at the outset.

Focusing. — Before focusing, the draw tube should be carefully extended to give the tube length for which the objective has been corrected by the maker.* This is not so important for low-power objectives but is essential for high powers if the optical efficiency is to be maintained. In the case of oil-immersion objectives a departure of 5 mm. from the standard tube length has a marked effect upon the quality of the image.

The first step in focusing is to rack the tube downward until the objective is within its prescribed working distance from the slide. Make it an unailing practice to do this with both eyes free to judge the position of the objective. There will then be no chance of ruining a preparation, or of damaging the lens. Then *focus upward* with the eye over the ocular, using the coarse adjustment. When the field is partially in focus, shift to the fine adjustment to bring out detail. A low magnification

* Bausch and Lomb, 160 mm.; Carl Zeiss, 160 mm.; Ernst Leitz, 170 mm.

facilitates orientation of a desired field. The multiple nosepiece then allows a quick shift to a higher-power objective.

Choice of Objectives and Eyepieces. — The purpose of the examination is best served by the proper combination of objective and eyepiece. A common mistake among beginners with the microscope is a desire to use high magnifications when low ones would be better. With large objects or organisms, it often happens that high magnifications so enlarge details as to make impossible a comprehensive view of the whole object. It should be remembered that increased magnification means restricted field, a matter for consideration when composite preparations are being viewed, such as the microscopic organisms contained in a sample of water. It is largely personal preference among microscopists that determines whether a high-power objective and a low-power eyepiece, or the reverse combination, should be used to secure a given magnification. For examination of water a 16-mm. (10 \times) objective and a 7.5 \times or 10 \times eyepiece give the best combination. Individual organisms are best studied with the same eyepieces and a 4-mm. (43 \times or 45 \times) objective.

It is a common fault among beginners to neglect the condition of lenses. Fumes, finger marks, immersion oil, and dust seriously interfere with clear magnification and the ability to locate objects in the field. All lens surfaces and the ocular micrometer should be kept clean by wiping with lens paper moistened with xylene. Ordinarily, it is unnecessary and inadvisable to unscrew the objective combinations. They should be cleaned from the outside.

In the making of photomicrographs, careful consideration should be given to the choice of the optical combination used. A good general rule is to attempt photographically only those reproductions that are successfully viewed with the eye when working with the microscope in the usual way. Large objects, particularly those with a considerable third dimension, cannot be photographed satisfactorily with high magnification because reproduction is limited to a shallow depth of focus. Likewise, objects not possessed of sharp outlines and details suffer further from such defects upon enlargement. Low and medium powers give the best results with the microscopic organisms found in water. For bacteria the highest powers are used.

The Huygenian eyepieces may be used for photomicrographic work but they have a restricted field of view as compared with the more highly corrected types such as the hyperplane. With low powers and large objects, eyepieces are often omitted. The usual achromatic objectives can be used with good results; but the apochromatic, more highly corrected types are better. The best optical equipment for photography is an apochromatic objective and a hyperplane, or a projection, eyepiece.

Illumination. — Proper illumination is essential for the demonstration of detail and structure and for safeguarding the eyesight of the worker. He should become familiar with all those parts of his instrument that regulate illumination and should know the effect of each.

The best source of light is daylight, but artificial sources may be used with good results if daylight glass is interposed between the source and

the microscope. The plane mirror does not increase the intensity of the light and should be used with daylight and with low-power objectives. The concave mirror intensifies illumination and so is useful with weak sources of illumination and with medium- and high-power objectives. If the condenser is employed, however, only the plane mirror should be used. In connection with the condenser it is well to remember that for the finest work it should not be used dry with high powers but should have on its upper surface a drop of immersion oil that is in contact with the lower side of the slide.

The substage diaphragms should be employed to give to the field an illumination that is not trying to the eyes either by reason of its intensity or of its failure to make outline and structure easily visible.

Coverglasses. — Whenever liquid preparations are examined they should have a coverglass over them in order to eliminate the disturbing influence of the spherical shape of the surface of the liquid and that of evaporation and streaming. No. 2 coverglass, having a thickness of 0.17 to 0.25 mm., is the size best suited to match the correction of objectives. A variation of .05 mm. from this thickness will obliterate fine structure with medium- and high-power dry objectives. With low powers the influence of the coverglass is negligible, as it is likewise within certain limits with immersion objectives. With the latter the immersion oil prevents refraction as the rays pass from coverglass to objective. No. 1 glass with a thickness of 0.13 to 0.17 mm. is often employed with immersion objectives in order to gain working distance.

Magnification. — Magnification is the ratio between the linear size of the object magnified and that of its visual image. It is expressed as the number of times the linear dimension is magnified and is indicated by the multiplication sign; thus $100\times$ signifies that the visual image has a diameter 100 times greater than the object.

TABLE 14
MAGNIFICATIONS WITH ACHROMATIC OBJECTIVES AND HUYGENIAN EYEPieces
Tube length = 160 mm.

Objectives		Eyepieces				
Magnification	Equiv. Focus in mm.	5	6.4	7.5	10	12.5
2	48	10	12.8	15	20	25
4	32	20	26	30	40	50
10	16	50	64	75	100	125
21	8	105	134	157	210	263
43	4L	215	276	320	430	537
45	4S	225	288	338	450	562
60	3	300	384	450	600	750
97	1.9	485	621	727	970	1212

To determine magnification, use is made of a stage, or objective, micrometer which is a glass slip with divisions accurately ruled upon it, generally 1 mm. divided into hundredths. The image of the micrometer scale is projected virtually upon a sheet of paper by means of the camera lucida and may be measured by dividers or by a millimeter scale while the observer is looking through the microscope. The distance covered on the scale divided by the value of the coincident divisions of the micrometer gives the magnification. If 15 mm. are covered by 3 divisions (.03 mm.) of the micrometer, the image of the latter is enlarged $15 \div .03 = 500$ times, $500 \times$.

Magnification may be increased by using (1) a higher-power objective, (2) a higher-power eyepiece, (3) a longer tube length of the microscope.

The exact magnification of any optical combination and tube length must be determined, as there is more or less variation in lenses and instruments. The preceding table gives the approximate values as put forth by the Bausch and Lomb Optical Co.

Measurement of Objects. — *Units.* — The unit of measurement in micrometry is the micron (*plural* microns, or micra), which is denoted by the Greek letter μ . Its linear value is one one-thousandth of a millimeter. For the estimation of microscopic organisms by the Sedgwick-Rafter method, either the cubic standard unit or the square standard unit is used. The former is a cube 20 microns on a side, the latter a square 20 microns on a side (see Chapter VI).

Methods of Measuring Objects. — There are two procedures in general use for the measurement of objects. One is by means of the camera lucida and a stage micrometer. The object is first drawn on paper or has its outlines indicated, is then removed from the stage, and a stage micrometer is substituted. The image of the latter is made to coincide with the outlines of the object already upon the paper, and the size is judged by the number of divisions of the micrometer that are covered. This procedure is suitable for measurement of coarser objects but is not as accurate as the next method to be described.

The second method makes use of an eyepiece micrometer which is a glass disk with an arbitrary scale ruled on it. The scale may take the form of recurring parallel lines with each tenth line accentuated in some way, or it may be a square divided into smaller squares, or any recurring geometric form. The disk is placed in the eyepiece upon the diaphragm, which is at the focal point of the eyepiece and in that plane into which the real image is projected by the objective (O_3 of Fig. 27). The lines of the eyepiece micrometer are, therefore, superimposed upon the image of the object under view and both are projected into the eye of the observer. If the value of the micrometer rulings is known it is then possible to measure the length and breadth of the object. By revolving the eyepiece the lines of the micrometer can be brought into any convenient position about the central axis.

Calibration of Eyepiece Micrometer. — The divisions on the micrometer have a different value with each optical combination used and with each change in tube length. The standard tube length can seldom be exactly adhered to if it is desired to assign a definite value, or a round

number value, to the divisions. For purposes of calibration an objective micrometer is placed on the stage and the image of its scale brought to a focus in the field of the microscope, the standard tube length being employed. The ruling of the eyepiece micrometer is then seen superimposed upon that of the objective micrometer. The left-hand extremity of the objective scale is made to coincide with that of the eyepiece scale, or with any subdivision thereof. The value of the eyepiece divisions can now be estimated from the number of divisions that they cover on the objective scale, each division of the latter usually being .01 mm. For example, if 5 divisions of the eyepiece micrometer cover 8 of the objective the value of the former is $.08 \div 5 = .016$ mm., or 16 microns. If a value of 15 microns is desired it becomes necessary to extend the tube length, thereby increasing the magnification and reducing the number of divisions covered on the objective micrometer to 7.5 or .075 mm. The exact tube length should be made a matter of record and should always be used subsequently with the given objective and eyepiece in order to retain the same eyepiece micrometer rating.

Measurement with Whipple Micrometer. — For enumeration of microscopic organisms in water, standard procedure calls for the Whipple micrometer, described on page 96, a 16-mm. objective and a $7.5\times$ eyepiece. The micrometer must first be calibrated, using such a tube length that the sides of the large square cover 1 mm. on the stage, and the sides of the smallest squares .02 mm., 20 microns. The area of the smallest square is then equivalent to one standard unit.

Enumeration of organisms in terms of the standard unit is equivalent to reporting the area of the masses, length times breadth, without consideration of the third dimension, or thickness. The advantages and deficiencies of this unit are pointed out in Chapter VI.

The use of the cubic standard unit calls for estimation of the thickness of organisms. As pointed out above, the exact determination of this third dimension, which is seldom made, requires that the microscope be equipped with a graduated micrometer head on the fine adjustment. This makes possible the calculation of the distance through which the tube moves in focusing the length of the third dimension.

There are several ways in which the value of the third dimension may be obtained:

1. By measuring directly the thickness of several representatives of a species with the Whipple micrometer and applying the average of the several measurements to all the organisms that are counted. This procedure is possible when the organism presents different views of its body.

2. By noting the geometric shape of the organism to be spherical, cylindrical, or ellipsoidal and then using the measured diameter of revolution of the sphere, cylinder, or ellipsoid as the third dimension. This is the most common method and entails no direct measurement of thickness.

3. By focusing upon the top and bottom, when the organism is transparent, reading the measurement upon the calibrated micrometer screw, and using this as the third dimension. When the organism is not trans-

parent it is possible with certain symmetrical shapes to focus on the periphery and top of the organism and thus to obtain half the thickness.

4. By manipulating the position of the organism, when its thickness cannot be measured by the foregoing methods, so that it presents a view of the third dimension that can be measured. Movement of the coverslip, or the use of a fine needle or stiff bristle will facilitate this shift in position.

Killing and Preservation of Microscopic Organisms. — For the technique of killing and preserving microscopic organisms, the reader is referred to works on histology and microscopical technique.

The microscopic organisms may be preserved in permanent mounts upon glass slips, but for practical study it is more convenient to preserve them in mass in 2-oz. bottles. The following killing and preservative fluids may be found useful:

King's Fluid (for preserving algæ, etc.). —

	grams
Camphor-water*	50
Distilled water	50
Glacial acetic acid	0.50
Copper nitrate, crystals	0.20
Copper chloride, crystals	0.20

Schaudinn's Solution (for fixation of protozoa). — Mixture of 2 parts of saturated aqueous sublimate (mercuric chloride) and 1 part of absolute alcohol. If desired a trace of acetic acid may be added.

Formaldehyde. — For killing, use a 40 per cent solution, sold under the name of "Formalin." For preserving, use solutions varying from 5 to 10 per cent, according to the organisms.

Picro-sulphuric Acid (for killing). —

Distilled water saturated with picric acid	100 cc.
Sulphuric acid, strong	2 cc.

After using, wash with 60 per cent alcohol.

Osmic Acid (for killing). — Expose the sample to be treated to the vapors from a 0.1-2 per cent solution.

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* Made by letting a lump of camphor stand in distilled water for a few days.

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CHAPTER VI

RECORDS OF EXAMINATION

Good statistical sense and acquaintance with statistical methods are the prime requisites for success in compiling records of catches, analyzing them, and generalizing from the results obtained. Well-conceived book-keeping methods will serve a number of useful purposes besides recording the condition of the sample at the time of analysis. They furnish in retrospect a picture of the biological changes in relation to the various factors that influence the growth of organisms, and so establish relative values in control. They also present information of benefit either in forecasting future conditions or in serving as a guide to methods of reservoir treatment, water purification, or stream regulation.

The water analyst deals commonly with two types of records, those referring to the plankton and those giving information about the larger aquatic organisms. Of these, the first is the more complex and important problem, particularly in relation to drinking water; it is, therefore, dealt with at length in this chapter. The second is so variable, depending upon the object and methods of study, that no standardized procedure has as yet been developed. Records of larger aquatic organisms are, therefore, taken up more directly in connection with such special studies as fall within the scope of this book. It stands to reason, however, that many of the methods of preparing and analyzing plankton records dealt with in this chapter are also applicable to studies of larger aquatic forms of life.

RECORDS OF PLANKTON CATCHES

The microscopical examination of water samples shows that the concentrate contains minute organisms of various kinds, fragments of larger animals and plants, masses of grayish or brownish flocculent material, and fine particles of inorganic matter.

The microscopic organisms vary in size and in their mode of occurrence. Some are found as separate individuals, others are joined together in filaments, or in masses or colonies; some are one-celled, others are many-celled; some are extremely simple in structure, others are

complex; some are scarcely larger than bacteria, others are easily visible to the naked eye. It is difficult to establish a satisfactory system for recording the presence of these varied forms, and a number of different methods are in use.

The inorganic or mineral matter included in the concentrate consists of fine particles of clay or silica and flocculent masses of iron or other chemical elements. It is usually not considered in microscopical reports; more information about it can be obtained by direct examination of the sediment and by methods of physical and chemical analysis. The masses of grayish or brownish flocculent material are usually of organic origin. They are called "amorphous matter" because of their formless nature. The term covers all the irregular masses of unidentifiable organic matter, both living and dead. It does not include vegetable fibers, vegetable tissues, or other débris; neither does it include mineral matter except when it is intimately mixed with the flocculent material. Living "amorphous matter" consists chiefly of bacterial slimes (zoöglœa) in which gelatinous masses of bacteria are attached to organic food substances or to mineral matter. The custom of estimating the quantity of amorphous matter in water was adopted in the belief that a record of its volume gave significant information in connection with the other analytical determinations. It is doubtful whether this is so, and the determination of "amorphous matter" is probably of value only when its origin is known and quantitative estimates of its presence are needed in comparing different samples of the same water. In the study of the self-purification of streams below sewage treatment plants, for example, this record may be significant.

Methods of Expressing Results. — In Chapters IV and V a number of methods of expressing the findings of the examination of plankton samples have been mentioned. The methods fall into two classes, bulk measurement and counting. The former is rough but rapid; the latter, refined but time-consuming. In the adoption of a method of expressing results the analyst should be guided by the nature of the problem in hand. Far too often tedious processes are employed when simpler methods would yield adequate results.

Bulk Measurement. — The results of bulk measurement are commonly expressed in cubic centimeters of plankton per cubic meter of water. This unit is equivalent to parts per million by volume, and since the specific gravity of most plankton is close to 1.0 it agrees well with the common method of expressing the results of a sanitary water analysis, namely, parts per million by weight.

In recording catches in terms of bulk, the method of concentration should always be specified.

Individual Counting. — The oldest system of reporting the varieties and quantities of organisms found in the microscopic examination of water is the individual count, i.e., the enumeration of the individual organisms. Owing to the variation in the modes of occurrence of the organisms, however, it is difficult to state what constitutes an individual organism. What shall be the unit: the cell or the filament, colony, or mass? Practice has varied. The Massachusetts State Board of Health many years ago adopted the following system. All diatoms, desmids, unicellular algæ, rhizopods, crustacea, and nearly all rotifera and infusoria are counted as cells; the filamentous algæ as filaments; the social forms of infusoria and rotifera as colonies; and many of the algæ that occur as irregular thalli, as masses.

This system, which, for convenience, may be called the "individual counting system" does not always give satisfactory results. It is often found that a sample which simple inspection shows to be heavily laden with algæ and which is offensive both in appearance and in odor gives a low count, whereas a sample which is clear in appearance and agreeable to the taste gives a very high figure. This is due largely to the great difference in the size and mode of occurrence of the organisms reported as individuals. A great mass of *Clathrocystis* is given no more weight than a tiny *Cyclotella*. Each counts as one, although the former may contain a thousand times as much organic matter as the latter.

Standard Unit. — In order to correlate the numerical result of the examination more closely with the actual character of the water and its physical and chemical analysis, the author devised a system of quantitative microscopy in which the size of the organisms is considered as well as the number of individuals. This system, known as the "standard unit method," was first developed in 1889 in order to obtain a quantitative estimate of the amount of amorphous matter in the concentrate. Later it was extended to organisms as well. Its use now is widespread.

The microscope, through the use of a calibrated ocular micrometer, lends itself readily to two-dimensional mensuration. It is a simple matter to determine the average diameter or length and breadth of the organisms under observation or to estimate their area directly. The actual determination of their third dimension is a more time-consuming matter. Most organisms rest in water in such a position that their greatest cross-sectional area is horizontal; only rarely are they seen in the reverse position. For this reason the unit of measurement adopted by the author was a unit of area rather than volume. The standard unit to which all measurements are referred is represented by the area of a square 20 microns on a side, i.e., by 400 square microns. This unit was chosen because, with the magnification ordinarily secured in count-

ing microorganisms, the estimation of their surface area is readily made.

The ocular micrometer used to make the measurements has been shown in Fig. 22. It is subdivided to correspond to the standard unit. The large square, which covers one square millimeter on the stage of the microscope, is divided into four equal squares. Each of these contains 25 smaller squares (squares of the second order) which in turn contain 25 standard unit squares (squares of the third order). The eye readily divides the sides of the squares of the second order into fifths, and this division gives the side of the standard unit square. As shown in the figure, it is possible mechanically to subdivide one of the squares of the second order further into squares which are the actual size of the standard unit.

The use of the standard unit system does not involve much additional labor in counting. Many organisms are so constant in size that they can be counted individually and then reduced to standard units by multiplying by a constant factor that expresses their size. Filamentous forms of constant width can be measured in length and then reduced to units by multiplying by the average diameter. Irregular masses and variable colonies can be estimated directly in units.

The unit system does not always give better results than the counting system, and it is sometimes advisable to state the results both in numbers of individuals and in standard units. The areal standard unit is the basis of stating most of the results in this book.

Cubic Standard Unit. — The most desirable standard unit of measurement for reporting microorganisms is a volumetric one. Accurate use of such a unit, however, entails a third measurement of the organisms, which, as pointed out in Chapter V, is usually difficult to secure. Estimation of the third dimension, unless done by a seasoned and patient observer, is likely to be unreliable.

The cubic standard unit adopted in "Standard Methods" (1925), is a volume with a thickness of 20 microns and of the same horizontal projection as the standard unit. Its volume is, therefore, $20^3 = 8000$ cubic microns. Since 1 cc. contains 10^{12} cubic microns, it follows that 10^6 cubic microns equal one part per million by volume. Now 10^6 cubic microns equal $\frac{10^6}{8 \times 10^3} = 125$ cubic standard units. To obtain parts per million by volume, therefore, divide the number of cubic standard units by 125.

The computation of cubic standard units from the individual measurements is facilitated by the use of a logarithmic chart, such as Fig. 31, or by a table, such as outlined in Table 15 and commonly included in most

engineers' handbooks. In Fig. 31, spherical and cubical volumes are given directly for any measured diameter or side; cylindrical volumes are stated in terms of the diameter of the circular cross-section and must be multiplied by the length of the cell or filament; ellipsoidal volumes are given in terms of the third diameter and must be multiplied by the product of the large and short diameters of the elliptical section observed. Conical volumes are obtained by taking one-third of the cylindrical ones, and wedge-shaped or pyramidal volumes by dividing the cubical volumes by two or three, respectively.

In practice the shapes of the different plankton organisms may be approximated by using one of the solids listed above or by a combination of several of them.

Purdy, who is responsible for the introduction of the cubic standard unit of 8000 cubic microns, illustrates its use in connection with a table of areas and volumes of common geometric figures, such as Table 15, as follows:

TABLE 15

AREA OF CIRCLE AND VOLUME OF SPHERE AND OF CUBE FOR A GIVEN DIAMETER

Diameter	Area of Circle	Volume of Sphere	Volume of Cube
0.5	0.196	0.065	0.125
1.0	0.785	0.524	1.000
2.0	3.142	4.189	8.000
3.0	7.069	14.137	27.000
4.0	12.566	33.510	64.000
5.0	19.635	65.450	125.000
Etc., up to 30 or 40			

(1) Suppose a *Volvox* sphere be found, measuring 22 linear standard units in diameter. A glance at the complete table shows that such an organism has a volume of 5575 cubic standard units.

(2) A tiny section of alga (cylindrical), one-half standard unit in diameter and 15 standard units long, has a volume of about 3 cubic standard units; that is, area of end (0.196) multiplied by length (15) equals 2.9.

(3) A flagellate (the cone-shaped *Euglena*, for example), whose larger end has a diameter of 2 standard units (linear) with a total length of 6 standard units, has a volume of about 6 cubic standard units. (Area of end multiplied by one-third the length.)

(4) A cyclops, whose general shape is that of a cone with rounded base, measures, say, 12 standard units (linear) in diameter at the point where the body begins to round off to form the "head end." From this point to the tip of the tail is, say, 22 standard units in length. The volume is therefore found by multiplying the area of the base by one-third the length, or $113 \times 7\frac{1}{3} = 829$. Add to this the volume of

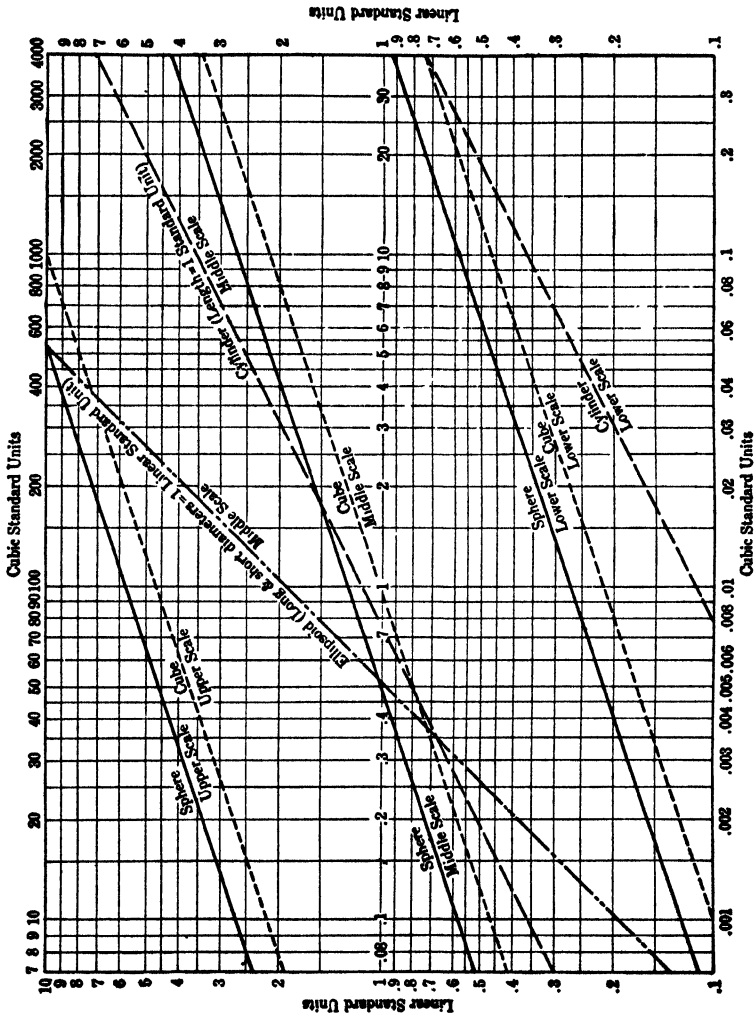


Fig. 31. — Chart for the Determination of Cubic Standard Units from the Linear Dimensions of Organisms.

the head end (one-half a sphere whose diameter is 12 linear standard units) and we have $829 + 452 = 1281$ cubic standard units as the approximate volume. If egg sacs are present they may be measured separately.

A cubic unit more consistent with the expansion of the metric system would be the cubic hecto-micron $[(100 \mu)^3]$, that is, a cube 100 microns on a side. The use of this unit in relation to 1 cc. of the sample would give parts per million by volume directly. Its size is indicated in the cube of the linear dimensions of the squares of the second order on the Whipple micrometer.

Organic Unit. — Juday has expressed the opinion that the future standard of comparing the microscopic load of water will be one based upon the chemical composition of the organisms. Each year the research of biologists presents the scientific world with new material in relation to the composition of microorganisms. At some future date, therefore, it will be possible to place quantitative microscopy on a chemically comparable basis. While this may be desirable from the standpoint of fish culture, the chemical composition of the organism is probably not as important in sanitary water analysis as is a numerical statement of the quantity and species of plankton present. The chemical composition of a large number of organisms is shown in Tables 30 to 32 (Chapter VIII).

Record Forms. — The results of analyses may be recorded in tabular form, graphically, photographically, or in other ways. Single analyses are commonly tabulated, while the results of series of examinations are frequently presented by graphical methods. The form adopted naturally depends upon the type of work in hand and upon the objects of the study.

Tabular Forms. — A form commonly used in recording the results of a single analysis by counting methods is shown on page 130. For purpose of illustration the results of examination of an impounded water are shown in this tabular form. The system of reporting is that of the areal standard unit.

In column (1) of this form are printed the names of the common organisms, grouped according to the system of classification employed in Part II, Determinative Microscopy. Several blank lines are provided below each group to permit a record of the less common organisms which may at times be present in large numbers.

Column (2) permits jotting down the dimensions of those organisms which are of uniform size in the sample and are therefore more readily counted by numbers. In counting filaments it is often convenient to jot down in column (2) the average diameter or cross-sectional area of the filament and to record the length only under (4). When this is done

it is well to indicate it by a multiplication sign following the measurement.

The numbers or units enumerated in the survey and total count* are recorded in columns (3) to (5). The ten numbered vertical columns under column (4) correspond to the ten squares commonly examined in the total count. Column (5) gives the sum of the values in the ten squares.

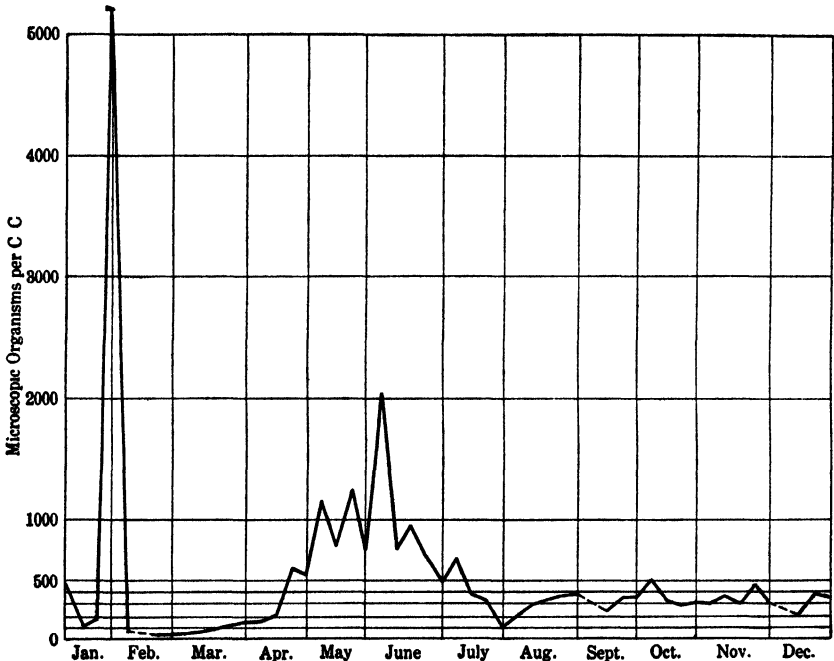


FIG. 32. — Graphic Record of Microscopic Organisms in Sudbury Reservoir, 1918. Arithmetic Plot.

The units per cubic centimeter are recorded in columns (6) and (7). The values in these columns are obtained by multiplying the figures in columns (3) and (5) by the survey and total count multipliers, respectively (see page 98) or, in case numbers or lengths of organisms have been recorded, by multiplying such numbers or lengths first by the average size of the organisms (column 2). The sum of columns (6) and (7) gives the total units of organisms in one cubic centimeter of the original sample.

Graphic Records. — The yearly and seasonal variation in microscopic counts can be compared in tabular or in graphic form. When a large number of results are to be studied, graphic methods of presentation are especially useful. The number or the standard unit value

For an explanation of the survey and total count see Chapter V.

MICROSCOPICAL EXAMINATION (Continued)

Organisms	Average Size (Stand. Units)	Number of Organisms or Standard Units											Standard Units per cc.		
		Sur- vey of Cell	Total Count of Fields										Sur- vey	Total Count	
			1	2	3	4	5	6	7	8	9	10			Total
(1)	(2)	(3)	(4)										(5)	(6)	(7)
FUNGI:															
Crenothrix.....	0.2×	3	...	4	...	4	...	5	5	...	21	...	8
Leptothrix.....
Sphaerotilus dichotomus....	0.1×	10	10	...	2
Mold hyphae.....	1.0	2	...	2	4	...	8
.....
PROTOZOA:															
Anthophysa.....
Ceratium.....	10 0	1	1	...	20
Cryptomonas.....	1.0	1	...	1	2	...	4
Dinobryon.....	0.5	10	8	18	...	18
Monas.....
Peridinium.....	6 0	...	1	2	1	...	1	5	...	60
Synura.....	10	10	20	...	40
Trachelomonas.....	1 0	...	1	1	1	1	...	2	6	...	12
Uroglenopsis.....
Vorticella.....
Codonella.....	8 0	1	1	...	18
Mallomonas.....	2.0	1	1	2	...	8
.....
ROTIFERA:															
Anurea.....	20.0	20	8	...
Polyarthra.....	25.0	24	12	...
Rotifer.....
.....
CRUSTACEA:															
Cyclops.....	...	500	10	...
Daphnia.....
.....
OTHER ORGANISMS:															
Anguillula.....	5.0	20	2	...
Sponge spicules.....	12.0	16	4	...
.....
TOTAL ORGANISMS													36	4571	
													4607		
AMORPHOUS MATTER													540		

REMARKS: Areal Standard Units are reported.

of organisms is best plotted vertically, the time element horizontally. The most common plot of this type employs arithmetic scales for both variables. When there are very large fluctuations in the number of organisms, however, it is sometimes better, in order to conserve space and present a better balanced plot, to use a logarithmic scale for the organisms. A graphic record of the number of organisms in the Sudbury Reservoir of the Metropolitan Water Supply of Boston for 1918

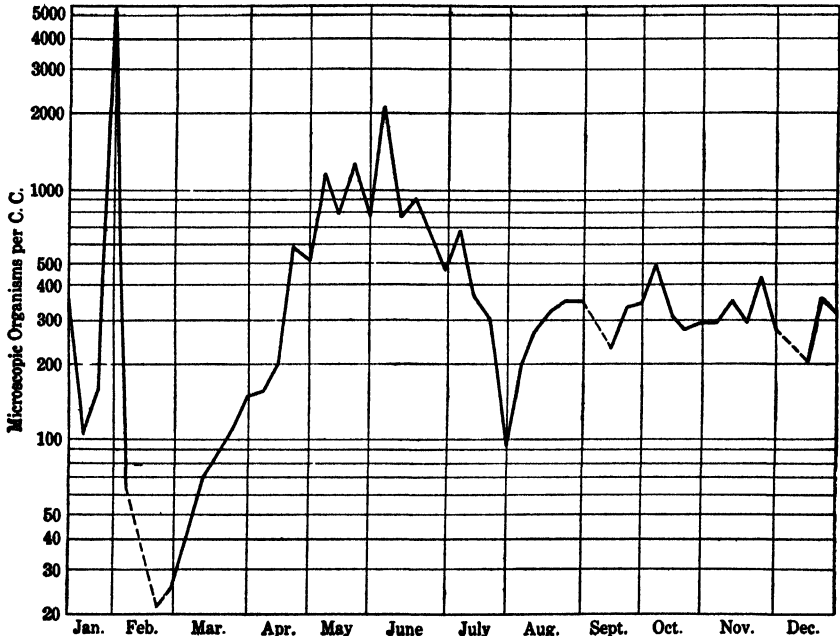


FIG. 33. — Graphic Record of Microscopic Organisms in Sudbury Reservoir, 1918. Semi-logarithmic Plot.

is shown in Figs. 32, 33, and 34. Figure 32 shows the plain arithmetic presentation, and Fig. 33 the semi-logarithmic one. In Fig. 34 the Lohmann method is utilized. The aim of this method is to present graphically those records that show the most extreme variations. For this purpose Lohmann takes the volume of a sphere as representing the number of organisms and plots the radius. The radius is given by the equation $R = \sqrt[3]{\frac{1}{4}V}$ where R = the radius and V = the number of organisms. The Lohmann method has been used extensively by limnologists. The three plots show the same results in approximately the same space, but leave very different visual impressions.

Photographic and Other Records. — Houston's method of preparing photomicrographs for purposes of recording the amount and kind of

suspended matter in water has been referred to in Chapter V. In his book, "Rivers as Sources of Water Supply," Houston states: "The photographs, it should be noted, are not taken for purposes of beauty, but in order to obtain pictorial, qualitative, quantitative, and comparative records of the varying condition of the river water throughout the year."

The use of cotton-disk records has been discussed in Chapter IV.

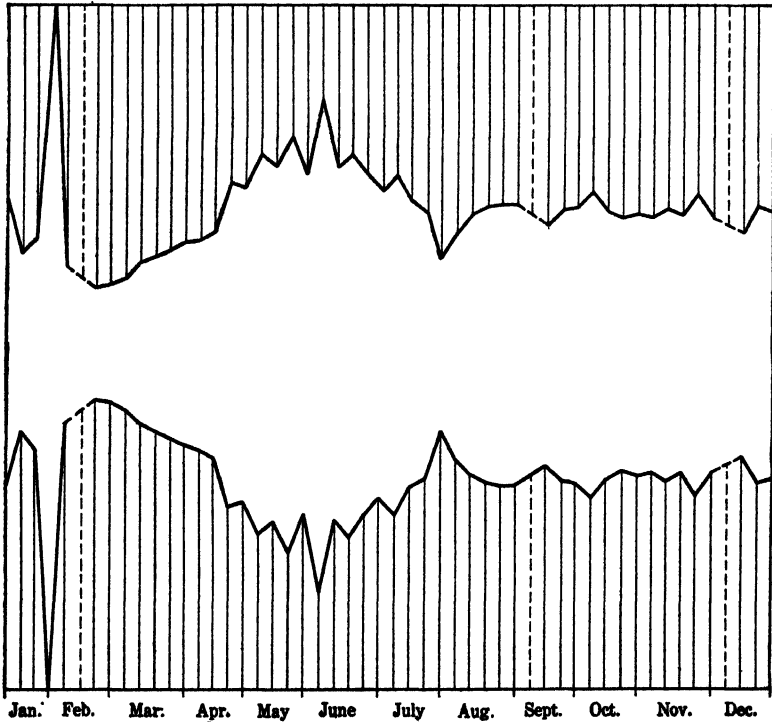


FIG. 34. — Graphic Record of Microscopic Organisms in Sudbury Reservoir, 1918. Lohmann Plot.

Analysis of Records. — The value of good records is greatly enhanced by thorough analysis of the findings. The intelligent study of such records is aided by thorough training in the use of statistical methods, adequate discussion of which falls beyond the scope of this book. The reader is referred to standard statistical texts for detailed information on this subject.

Measures of Central Tendency and Variation. — A few statistical characteristics of microscopic analysis will be briefly mentioned. One of these is the variation of microscopic counts arranged in order of magnitude. As shown in Fig. 35, such a cumulative array, as exempli-

fied by the Sudbury records for 1918, plots close to a straight line on logarithmic probability paper. This signifies that variations from the mean are geometric rather than arithmetic and that counts several times as small or as great as the geometric mean occur relatively frequently. In our example, values larger than twice the geometric mean or smaller than one-half the geometric mean are found half the time,

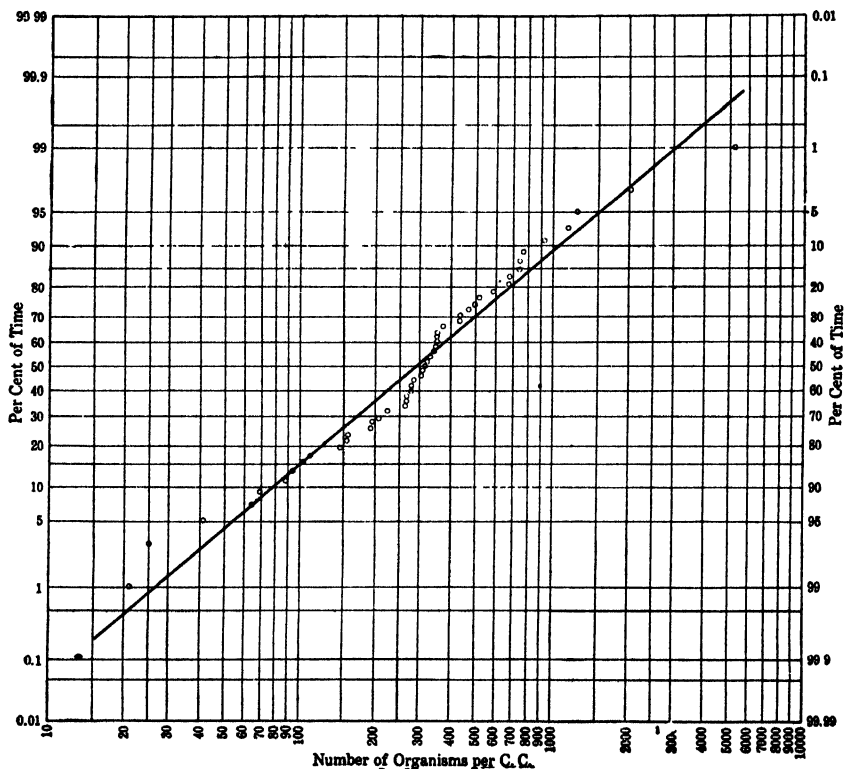


FIG. 35. Cumulative Frequency Distribution of Microscopic Organisms in Sudbury Reservoir, 1918. Weekly Analyses Plotted in Order of Magnitude on Logarithmic Probability Paper.

i.e., they are just as likely to be observed as not. Counts greater than five times the geometric mean may be expected five per cent of the time, and counts less than one-fifth the geometric mean likewise. The parameters or yardsticks that best express a series of this type for purposes of generalization and comparison are probably the geometric mean and the standard geometric deviation, equal in our case to 290 and 2.8, respectively. These two measures, one of central tendency, the other of dispersion, furnish the statistically initiated with as much information

as can otherwise be obtained from a detailed interpretation of the graph or of the complete series of figures. The use of measures of this type, however, implies a familiarity with biometric principles not frequently possessed by water works operators. In a study made in 1906 of 66 lakes and reservoirs in New England, the author therefore devised an "index of frequency" which serves as a parameter of the frequency of occurrence of microscopic organisms and obviates some of the difficulties arising from a lack of statistical appreciation.

Index of Frequency. — For purposes of comparison, the frequency of occurrence of microscopic organisms and the intensity of their growth were used to obtain for each reservoir a single figure which would best represent the relative amount of trouble caused by organisms. This figure, or parameter, was called the "index of frequency" and calculated as follows:

It was assumed in accordance with experience that when the organisms were less in number than 500 per cc. they would cause no trouble; between 500 and 1000 per cc., little trouble; between 1000 and 2000, noticeable trouble; between 2000 and 3000, decided trouble; and that above 3000 the trouble would be serious. From the records of analyses, the time during which organisms were growing within each one of these limits was ascertained. For purposes of comparison, the time was then expressed in per cent of the total period of observation. These time ratios were then weighted as follows and added together: for numbers between 500 and 1000, one-half the per cent of time; for numbers between 1000 and 2000, the per cent as computed; for numbers between 2000 and 3000, twice the per cent; and for numbers above 3000, three times the per cent. Organisms of all kinds were considered, without regard to genera. While this over-values the effect of certain organisms and under-values that of others, it is believed that the grouping is generally fair. The method of computing the index of frequency is illustrated in Table 16 for the weekly records of the Sudbury Reservoir in 1918.

To appreciate the significance of the index of frequency, it is necessary to become familiar with the information implied by the various possible values that the index may assume. Thus an index of 50 means either that the growth of organisms was noticeable half the time, or that, if noticeable for less than half the time, organisms were more troublesome during the period of their presence.

The maximum value that the index of frequency can attain is 300, but it is rare indeed that any natural water gives an index of more than 100, and in the best stored waters the index is generally less than 10. The minimum of course is 0.

TABLE 16
COMPUTATION OF INDEX OF FREQUENCY
SUDBURY RESERVOIR, 1918

Number of Organisms per cc.	Number of Weeks when Organisms Were Found between the Given Limits	Per Cent of Time	Per Cent Weighted
0-500	37	76	$0 \times 76 = 0$
500-1000	8	16	$\frac{1}{2} \times 16 = 8$
1000-2000	2	4	$1 \times 4 = 4$
2000-3000	1	2	$2 \times 2 = 4$
3000-	1	2	$3 \times 2 = 6$
	—	—	—
	49	100	22
			Index of Frequency

The following figures show the numbers of organisms found in three groups of lakes classified according to the index of frequency.

TABLE 17
LAKES CLASSIFIED ACCORDING TO THE INDEX OF FREQUENCY
1900

	Group I	Group II	Group III
Number of Lakes and Reservoirs in the group..	28	18	20
Limits of Frequency Index.....	0-25	25-50	50-100
Average index of frequency.....	12	39	78
Organisms per cc. mean yearly average.....	362	776	1410
Organisms per cc., minimum yearly average....	54	441	984
Organisms per cc., maximum yearly average....	1413	2800	3090
Organisms per cc., mean average for 4 summer months.....	414	1023	1965
Organisms per cc., minimum average for 4 summer months.....	66	227	985
Organisms per cc., maximum average for 4 summer months.....	1058	4588	7659

Measures of Fluctuation. — The parameters so far suggested apply to records of examination arrayed in order of magnitude. Their use gives information relating to the per cent of time during which growths of a given amount have been recorded or may be expected. In water

works management, particularly in water purification, it is valuable to know also how the growths fluctuate from day to day, week to week, etc. This entails an analysis of the records arrayed in order of time (see Fig. 32).

Several parameters have been suggested as expressing variation in time or fluctuation. One of the best so far devised is "Crum's coefficient of fluctuation." * The theory and computation of this parameter will not be explained here. Parameters of fluctuation can be used to advantage in giving information as to the behavior of various genera of organisms in different waters and at different seasons of the year. Expressing as they do in a single figure the many changes that occur in different intervals of time, their use provides a basis for the comparison of otherwise complex conditions. Measures of fluctuation are particularly valuable in the study of the success of algæ control and of the microscopic loads with which purification plants may have to cope.

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CHAPTER VII

LIMNOLOGY — PHYSICAL CONDITIONS

In order to interpret correctly the occurrence and distribution of microscopic organisms in water it is necessary first to understand those environmental factors that influence most their genesis, growth, and decay. Since standing water presents a somewhat different environment from running water, it is customary to consider separately lakes and ponds, and rivers and brooks. That branch of science which deals with lakes and ponds—their geology, physiography, physics, chemistry, and biology—is called *limnology*; that which considers streams and brooks is known as *rheology*. The roots of these words are derived respectively from the Greek words “limnos,” a lake, and “rheos,” a stream.

Limnology includes a great variety of subjects that aid but little in the interpretation of the microscopy of drinking water. For this reason only such limnological work as is closely related to microscopic organisms is considered in this and the two succeeding chapters. The most important limnological conditions may be classified as follows:

1. Physical conditions: Temperature, density, viscosity, transparency, and movement of the water. These are considered in the present chapter.

2. Chemical conditions: Dissolved gases, inorganic and organic food materials. For a discussion of these factors see Chapter VIII.

3. Biological conditions: Seasonal distribution, dispersion, and behaviorism. These are subjects that are dealt with in Chapter IX.

Besides these conditions, the location of lakes, their shape, size, and depth, the source of their waters, the character of the watershed, and the meteorology of the region, are all of importance to the organisms living in water, but they can be considered only incidentally.

Most of the conditions named are mutually dependent and combine in some measure to contribute to the three essentials of plankton existence, namely, food, heat, and light. The physical factor of temperature, for example, stimulates plankton activity directly by developing conditions of warmth essential to reproduction and growth. It also acts indirectly through its influence upon water density and viscosity to produce water movements and environmental conditions that determine the horizontal and vertical distribution of the plankton and carry supplies of food to the growing organisms. In studying the various factors, therefore, this interrelationship must not be lost from view.

HEAT CONDITIONS

Physical Properties of Water. — Some of the thermal properties of water make it an ideal environment for the development of primitive forms of life. Thus, water has a high specific heat, which means that much heat is required to warm it and likewise much cold to cool it. The specific heat of water is five times as great as that of air, and the variation in the temperature of lakes and ponds during the various seasons of the year, therefore, is far less than in the overlying atmosphere. The greatest variation naturally occurs at the surface of the water as the lower layers are dependent upon conduction, convection, and radiation for changes in temperature. Water is a poor heat conductor and opposes the flow of heat through it. It also has only a slight power of diathermancy, i.e., it permits the passage of radiant heat to a limited degree.

Diathermancy. — Forel experimented on the diathermancy of water by comparing the readings of thermometers with blackened and with ordinary bulbs at a depth of 1 meter. He obtained the results found in Table 18.

TABLE 18
TEMPERATURE OBSERVATIONS ILLUSTRATING DIATHERMANCY
After Forel

Date	Time of Exposure (Hours)	Temperature of Water (°F.)	Excess in Temperature of Black Bulb Thermometer (°F.)
Mar. 27, 1871	10	44.4	10.8
July 25, 1873	17	72.0	14.0
July 26, 1873	15	74.3	15.3
Aug. 1, 1873	12	75.2	7.6

On the other hand, the transmission of heat through water by convection, that is, by motion of the water itself as a result of warming or cooling, is considerable. This takes place whenever the changes in temperature cause differences in density that operate to produce motion, i.e., rising or falling of the water strata, or when wind action forces warm water beneath colder layers.

Density. — The density of water depends chiefly upon its temperature; it is only affected to a slight extent by pressure and dissolved substances. The compressibility of water is very low. At the maxi-

imum density of water the coefficient of compressibility is only about .00005 for each atmosphere of added pressure. This means that at a depth of 339 feet (10 atmospheres) the density is increased from unity to 1.0005. Dissolved substances increase the density of water in accordance with their concentration and their own specific gravity. In natural fresh waters the variation in density due to dissolved substances is very small.

Whereas pressure and dissolved substances thus have but little effect upon the density of water in lakes and ponds, the variation in density due to temperature changes is relatively large. This variation becomes of great moment in explaining the stratification as well as the movements of water which are responsible for the distribution of plankton and, at certain seasons of the year, for the movements of food supply that result in large plankton growths.

Water attains its maximum density at 4° C. or 39.2° F. If its density at 4° C. is called unity, its density at other temperatures is given by the figures in Table 21, page 164. It is seen that the density of the water decreases when the temperature sinks below 4° C. as well as when it rises above 4° C. Water freezes at 0° C. or 32° F. Its density at this temperature is almost as low as at 10° C. In freezing, water expands about 8 per cent and the resulting density of the ice formed is about .917 of the density of water at 0° C. This difference in specific gravity causes ice to float. The specific heat of ice is about one-half that of water and its thermal conductivity is almost twice as great. This might lead one to believe that ice increases heat losses from the warm water to the cold atmosphere; but this is not so. Conduction of heat is extremely slow, and the sheet of ice formed on the surface of water prevents direct contact between air and water and inhibits heat radiation from the water into the atmosphere. The ice cover, therefore, actually reduces heat loss from the water to that of conduction through the mass of ice.

The variation in the density of water has but little direct effect upon microscopic organisms, whose structure seems to adapt them readily to changes in pressure. They exist with equal facility at various depths in ponds and lakes, those organisms that have powers of locomotion migrating readily from great to shallow depths and *vice versa* during short intervals of time. The viscosity of water, on the other hand, seems to have an important direct effect on microorganisms as it changes their flotation.

Viscosity. — Viscosity is the transient resistance of a fluid to deformation; it is internal friction. Any particle of higher specific gravity than water, suspended in water, tends to sink. This tendency is op-

posed, however, by the viscosity of the water. The more viscous the water the greater this opposition.

The viscosity of water varies greatly with the temperature. It is twice as great near the freezing point as at ordinary summer temperatures. Its influence upon the flotation of organisms is therefore large. The variation of viscosity is shown in Table 19.

TABLE 19
VISCOSITY OF DISTILLED WATER AT DIFFERENT TEMPERATURES

Temperature (°C.)	Viscosity Coefficient (Dynes per Square Centimeter)	Percentage Ratio to Viscosity at 0° C.
0	0.017780	100.0
5	0.015095	84.9
10	0.013025	73.2
15	0.011425	64.2
20	0.010015	56.3
25	0.008910	50.1
30	0.007975	44.8
35	0.007200	40.5
40	0.006535	36.8

Hazen has expressed this variation by the following approximate relationships:

$$v_t = v_{50} \frac{60}{t + 10}, \quad \text{where}$$

v_t = viscosity at any natural water temperature,

v_{50} = viscosity at 50° F.,

t = water temperature in ° F.

Summarizing the heat conditions of water, it becomes apparent that the temperature of water is indirectly also a measure of density, viscosity, and water movement, and will therefore furnish valuable information regarding the occurrence and distribution of microorganisms. Temperature measurements form an indispensable part of limnology.

Lake Thermometry. — Observation of the temperature of the water at the surface of a lake is a comparatively simple matter. All that is needed is an accurate thermometer in the hands of a careful observer. In smooth water the thermometer bulb is immersed in an inclined position just beneath the surface, and the reading is taken without removing it from the water. Parallax should be avoided by holding the ther-

nometer exactly at right angles to the line of sight. When the water is too rough for direct reading, some of the surface water is dipped up and its temperature ascertained. Thermometers provided with a cup in which the bulb is immersed have been constructed for this purpose, but direct observations are much to be preferred.

A "chemical thermometer" about 9 inches long, with a cylindrical bulb and stem graduated from 0° to 40° C. to the nearest fifth of a degree, is a convenient instrument for general use. If a Fahrenheit scale is used, it should read from 20° to 120° to the nearest half-degree. To protect the thermometer against breakage, it should be mounted in a wooden or metallic case, as shown in Fig. 36. The case can be weighted for use in obtaining sub-surface temperatures.

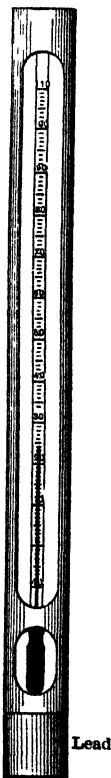


FIG. 36. —
Weighted
Thermom-
eter Case.

Sub-surface Temperatures. — The observation of the temperature of the water at depths below the surface is more difficult.

The readiest method for reasonably accurate results is to enclose a weighted thermometer in a stoppered bottle which is lowered to the proper depth and filled by withdrawing the stopper. After sufficient time has been allowed for the apparatus and thermometer to acquire the temperature of the surrounding water, the bottle is hauled to the surface and the reading taken without removing the thermometer from the bottle. If the bottle is sufficiently large, remains down long enough, and is then drawn rapidly to the surface where the thermometer is read immediately, the error should not exceed half a degree centigrade. This method becomes impracticable for depths greater than about 50 feet, when the use of some form of deep-sea thermometer becomes necessary.

There are many types of deep-sea thermometers. The instrument most widely used in oceanography is a special mercurial thermometer, such as that made by Negretti and Zambra of London, England (Fig. 37). The capillary of this instrument is constricted near the bulb and causes the mercury to separate when the instrument is inverted.

The shape of the constriction is such that no subsequent change in temperature is recorded (Fig. 37a). In operation the thermometer is lowered to the required depth and reversed as shown in Fig. 37c. The thermometer may also be inverted by a mechanism activated by a messenger (Fig. 37b) or by a propeller that spins when the instrument is pulled up a short distance. The thermometer must be hauled to

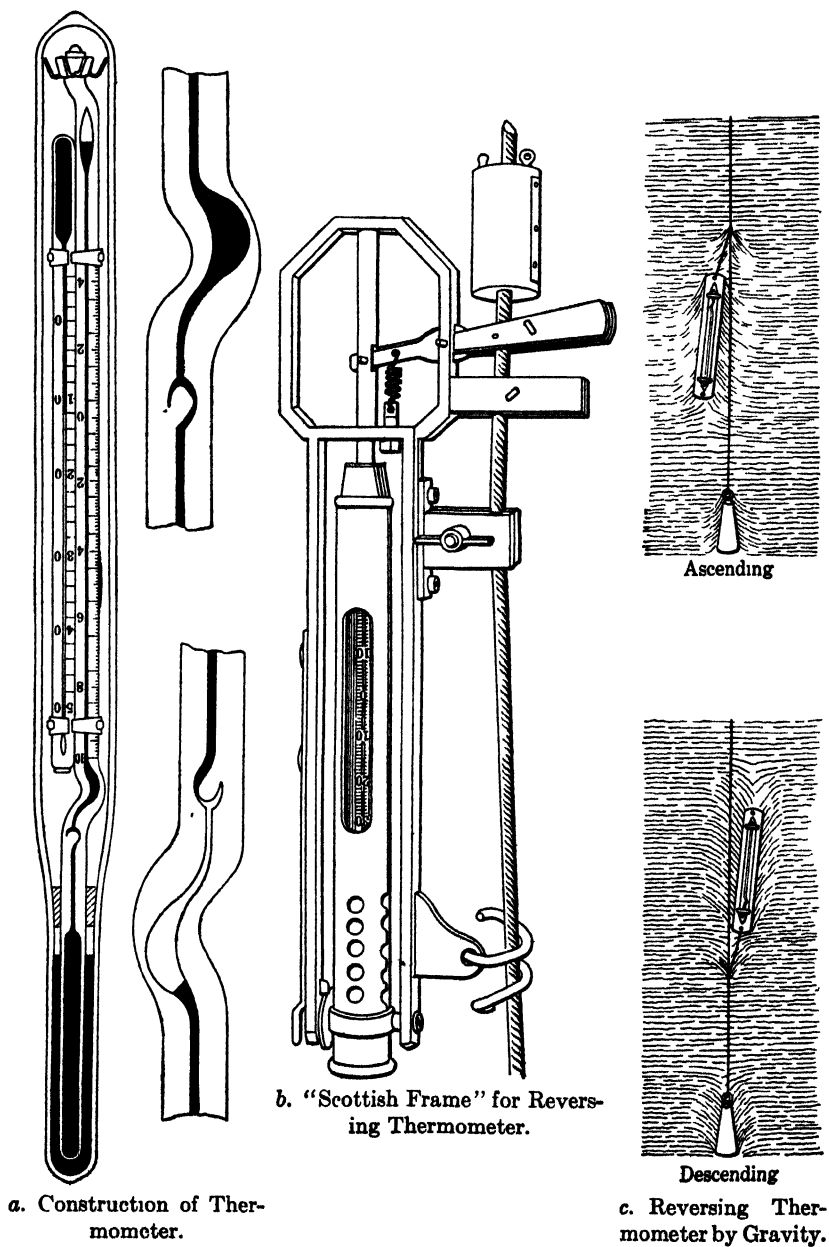


FIG. 37 *a, b, c.* — Negretti and Zambra's Deep Sea Thermometer.

the surface for each reading. For measurements at very great depths, Clark devised a thermograph in which the readings of a mercurial thermometer are recorded on a photographic film illuminated by an electric light.

Several forms of electrical thermometers have been suggested. Of these the one invented by H. E. Warren and the author, known as the "thermophone," has found wide application in limnological studies.

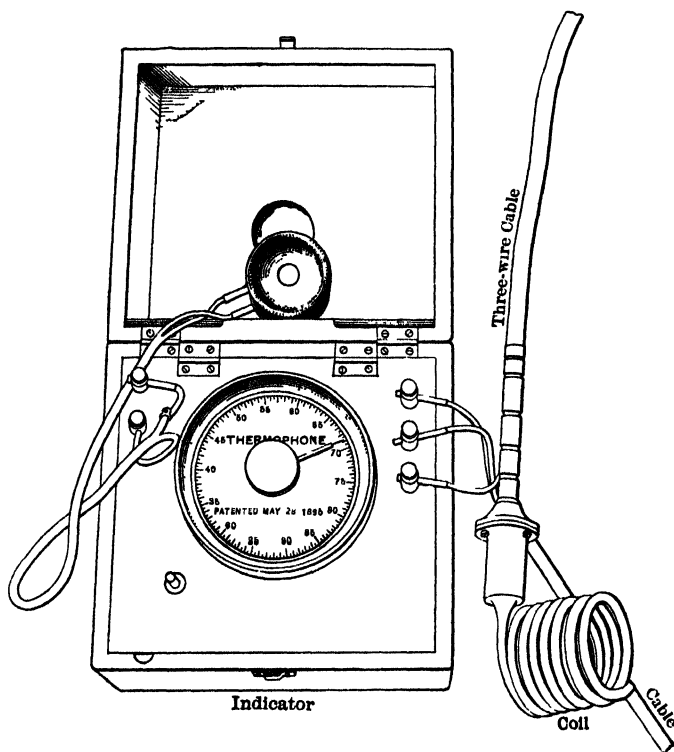


FIG. 38. — Thermophone.

For very fine measurements Barnes has devised a serviceable instrument in which temperatures are determined by changes in the electrical resistance of a platinum wire.

The Thermophone. — The thermophone (see Fig. 38) is an electrical thermometer of the resistance type. It is based upon the principle that the resistance of an electrical conductor changes with its temperature and that the rate of change is different for the various metals. Two resistance coils of metals that have significantly different electrical temperature coefficients, such as copper and German silver, are connected into adjacent arms of a Wheatstone bridge and are lowered into

the water to a stratum where the temperature is to be determined. The two coils are joined together at one end, their free ends being connected by lead wires to the terminals of a slide wire which forms a part of the indicator. A third lead wire extends from the junction of the two coils to a movable contact on the slide wire, having in its circuit a telephone and a current interrupter, the latter operated by an independent battery connection. The telephone and interrupter serve in place of a galvanometer to detect the presence of a current. The slide wire is wound around the periphery of a mahogany disk, above which there is a dial graduated in degrees of temperature. The movable contact which bears on the slide wire is attached to a radial arm placed directly under the dial hand, the two being moved together by turning an ebonite knob in the center of the dial. This indicator is enclosed in a brass case and is placed in a box which also contains the batteries. The sensitive coils are enclosed in a brass tube of small diameter which is filled with oil, hermetically sealed, and coiled into a helix. Connections with the lead wires are made in an enlargement at one end. The lead wires form a three-wire cable. The temperature of the lead wires does not affect the reading of the instrument because two of them are of low resistance and are on opposite sides of the Wheatstone bridge. They neutralize each other. The third lead wire is connected with the galvanometer and does not enter the equation. The readings of the instrument are independent of pressure. A wiring diagram of the thermophone is shown in Fig. 39. *A* and *B* are the coils of different metals. *CD* is the circular slide wire to which the coils are connected by means of lead wires *L* and *L'*. The two ends of *CD* are placed in the circuit of the battery, *M*. *G* is the galvanometer or current interrupter and telephone. It connects through the third lead wire with the junction of *A* and *B* and by means of the movable contact, *Y*, with the slide wire. The galvanometer will indicate zero current in the third lead

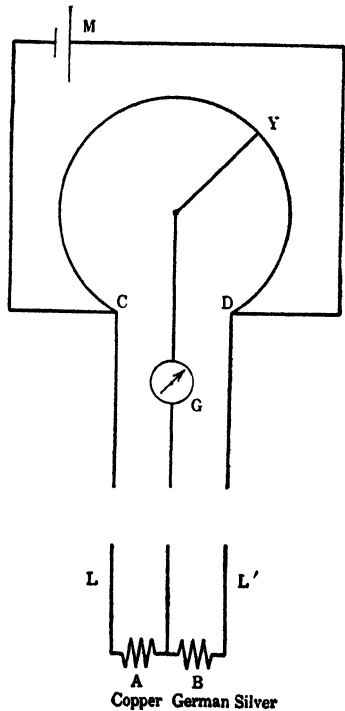


FIG. 39. — Wiring Diagram of Thermophone Circuit.

wire when the resistances are proportioned as follows: $\frac{A}{B} = \frac{CY}{DY}$. Since the resistances of *A* and *B* vary unequally with changing temperatures, each temperature will yield a different value of $\frac{A}{B}$. This value, measured by $\frac{CY}{DY}$ and expressed as temperature, is indicated by the position on the slide wire of the movable contact, *Y*. Equipping *Y* with a pointer, and the arc through which it moves with a temperature scale, completes the instrument.

The thermophone is operated as follows: The coil is lowered to the desired depth, the three lead wires are connected to the proper binding-posts of the indicator box, the current from the battery is turned on, the telephone is held to the ear, and the hand moved back and forth over the dial. A buzzing sound is heard in the telephone, increasing or diminishing as the dial hand is made to approach or recede from a certain point on the dial. At this point there is perfect silence in the telephone, and the hand indicates the temperature of the distant coil. With thermophones designed for atmospheric range, i.e., from -15° to 115° F., readings correct to 0.1° F. can be made. With a smaller range greater sensitiveness can be obtained. It is possible to construct thermophones that will read to thousandths of a degree.

Because of its accuracy, the ease with which the coil is lowered to any depth from the surface to the bottom of a lake, its extreme sensitiveness and rapidity of setting (one minute is sufficient), and its portability, the thermophone is adapted better than any other instrument to

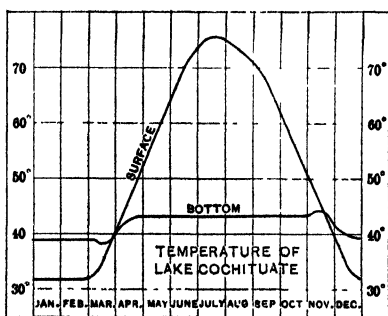


FIG. 40. — Seasonal Temperature Changes at Surface and Bottom of Lake Cochituate.

Lake Cochituate. The curves are based on a seven-year series of weekly observations. For the sake of simplifying the graph, the observations have been smoothed.

taking series of temperature observations in lakes at various depths. It has been used for this purpose to depths as great as 400 feet, and was employed at much greater depths by Prof. A. E. Burton, in Greenland, for obtaining temperatures in the crevasses of glaciers.

Temperature Changes in a Lake.

— The general character of the temperature changes that take place in a body of water are illustrated by Fig. 40, which shows the temperatures at the surface and bottom of

On tracing the line of surface temperatures, one observes that during the winter the water immediately under the ice stands substantially at 32° F., although the ice itself often becomes much colder than 32° at its upper surface. As soon as the ice breaks up in the spring the temperature of the water begins to rise. This increase continues with some fluctuations until about the first of August. Cooling then begins and continues regularly through the autumn until the lake freezes in December. If this curve of surface temperature were compared with the mean temperature of the atmosphere for the same period, a striking correlation would be noticed, and it would be seen that the water temperature is usually the higher of the two. When the surface is frozen there is no relation between air and water temperatures. During the spring and early summer, when the temperature of the water is rising, the water is but slightly warmer than the air,* but during the late summer and autumn it is about 5° F. warmer. The surface temperature of the water fluctuates with the air temperature during the course of the day as well as from day to day. The maximum is usually reached between 2 and 4 P.M. and the minimum between 5 and 7 A.M. The daily range is seldom greater than 5° F., though it may be much more. At the latitude of Boston the maximum surface temperature of the water of lakes during the summer is seldom above 80° F.†

In small shallow ponds the surface temperature follows the atmospheric temperature much more closely than in large deep lakes where the water circulates to considerable depths. In the latter the surface temperature is often below the mean atmospheric temperature during the early part of the summer, and occasionally during the entire summer.

Returning to the Lake Cochituate curves, Lake Cochituate is 60 feet deep. The temperature at the bottom during the winter, when the surface is frozen, is not far from that of maximum density (39.2° F.).

* It must be understood that it is the mean temperature of the air during 24 hours that is referred to, and not the maximum temperature during the daytime.

† A surface temperature of 92° F. was observed by the author at Chestnut Hill Reservoir on Aug. 12, 1896, at 3 P.M., after a week of excessively hot weather, during which the maximum daily temperature remained above 90°, while the humidity varied from 62% to 95%. At the time of the observation the air temperature was 95° and the humidity 70%. The temperatures of the water below the surface were as follows:

Surface.....	92.0°	10 ft.....	76.2°
1 ft.....	91.5	15 "	74.0
2 "	89.2	20 "	65.7
3 "	85.6	25 "	54.5
4 "	80.2	27 "	53.1
5 "	79.0		

The heaviest water is at the bottom; the lightest is at the top; the intermediate layers are arranged in the order of their density. Under these conditions the water is in comparatively stable equilibrium, but is *inversely* stratified, i.e., the colder water overlies the warmer water. This is the period of "winter stagnation."

As soon as the ice has broken up in the spring the surface water begins to grow warmer. Until it reaches the temperature of maximum density it also becomes denser and tends to sink. Thus, until the water throughout the vertical has acquired the temperature of maximum density there are conditions of unstable equilibrium caused by diurnal fluctuations of temperature that result in thorough mixing of all the water in the lake. These conditions, together with the mechanical effect of the wind, usually cause a slight temporary lowering of the bottom temperature at this season. Finally the temperature, and with it the density, throughout the vertical becomes practically uniform, and vertical currents are easily produced by slight changes in the temperature of the water at the surface and by the mechanical effect of the wind. This is the period of "spring circulation," or the "spring overturning." It lasts several weeks, but varies in length during different years.

As the season advances the water at the surface becomes warmer and lighter than that at the bottom, and finally the difference becomes so great that the diurnal fluctuation of surface temperature and the effect of the wind are no longer able to keep up the circulation. Consequently the bottom temperature ceases to rise, the water becomes *directly* stratified, and the lake enters upon the period of "summer stagnation." During this period, which extends from April to November, the bottom temperature remains almost constant and the water below a depth of about 25 feet is nearly stagnant.

In the autumn the surface cools and the water becomes stirred to greater and greater depths, until finally the "great overturning" or "fall overturning" takes place and all the water is in circulation. At this time there is a slight increase in the bottom temperature that corresponds to the temporary lowering of the temperature in the spring. Then follows the period of "autumnal circulation," during which the surface and bottom strata have substantially the same temperature. In December the lake freezes and "winter stagnation" begins.

Winter Conditions. — In a frozen lake the water in contact with the bottom of the ice stands always at 32° F. The temperature at the lake bottom varies with the depth and with the meteorological conditions obtaining at the time of freezing. In most lakes, and particularly in deep lakes, it stands at the point of maximum density; in shallow lakes it may be lower than that; under abnormal conditions,

it may be slightly higher. During the period of winter stagnation the bottom temperature sometimes rises very slightly on account of direct heating by the sun's rays. This is due to the diathermancy of the water. The variation in the temperature of the water between the surface and the bottom is illustrated in Fig. 41.

The cold water is usually confined to a layer, seldom more than 5 or 10 feet thick, in contact with the ice. Below this layer, as shown by the Lake Cochituate curve, the temperature changes but little to the bottom. This condition and the peculiar change in the curve at the bottom may be explained as follows: During the period of autumnal circulation the temperature is uniform throughout the vertical. As the weather gets colder the temperature throughout the vertical drops. Until the temperature has reached the point of maximum density the circulation of the water through the vertical takes place in part by thermal convection; below that temperature it takes place chiefly by wind action. If the wind is not sufficiently strong to induce complete circulation the bottom temperature ceases to fall at 39.2° F. Thus the bottom temperature at Lake Cochituate in December, 1894, was left at that point. Later the wind stirred the water to a depth of 45 feet, and above that depth the temperature became uniform at about 38.5° F.

Freezing usually occurs on a cool, still night. The surface water cools and freezes before the wind has had a chance to mix it with the warmer water below. The suddenness with which a lake freezes and the intensity of the wind prior to freezing determine the depth of the layer of cold water. The temperature of the air and the intensity of the wind previous to the time of freezing determine the temperature of the

water at the bottom. The Lake Winnepesaukee curve (Fig. 41) represents the effect of a current flowing between two islands. A layer of cold water about 18 feet thick was flowing over a quiet body of warmer water. The dividing line, at a depth of about 20 feet, is very sharply

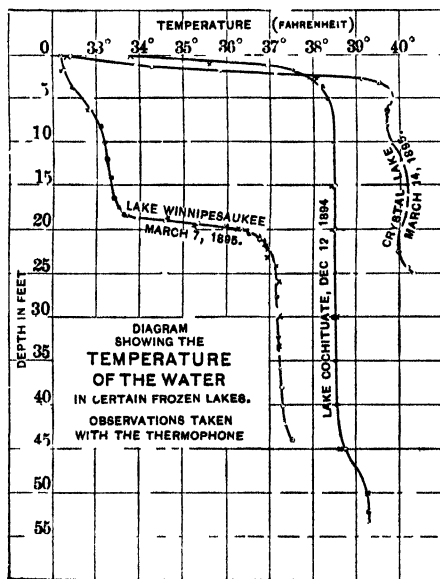


FIG. 41. — Vertical Temperature Gradients in Frozen Lakes. After FitzGerald.

defined. The Crystal Lake curve (Fig. 41) shows abnormal conditions produced by springs at the bottom of the lake.

Summer Conditions. — During the summer the temperature of the water is similarly affected by meteorological conditions. After the ice has broken up, the temperature of the water at all depths rises. Above 39.2° circulation takes place chiefly by wind action. If there were no wind, or if the wind were not sufficiently strong, the temperature at the bottom would not rise above 39.2°. In very deep lakes this happens, but in most lakes wind action causes it to rise somewhat above this point. It continues to rise as long as the difference in density between the water at the surface and at the bottom does not become too great for the wind to keep up circulation. In Lake Cochituate this difference of density is produced by a difference of about 5° in temperature. When stagnation has once begun the temperature at the bottom changes very little during the summer. It sometimes rises slightly on account of direct heating, as it does in winter. If warm weather occurs early and suddenly in the spring the required difference of temperature between the upper and lower layers is soon established, and consequently the temperature at the bottom remains low through the summer. If, on the other hand, the season advances slowly, the bottom temperature becomes fixed at a higher point. In Lake Cochituate the bottom temperature varies in different years from 42° to 45°.

The temperatures of the water between the surface and bottom during the summer may be illustrated by the two typical curves of

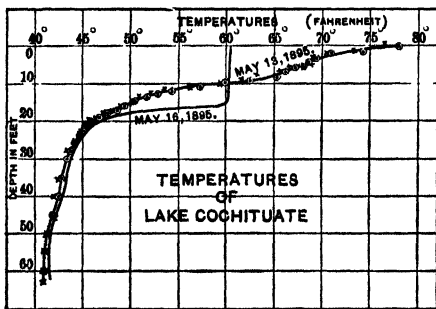


FIG. 42. — Vertical Temperature Gradients of Lake Cochituate during Warm Weather.

Fig. 42. Previous to May 13, 1895, the season had progressed gradually. On that day the atmospheric temperature rose to 90° and there was little wind. These conditions produced a uniform curve. Then followed several days of cold, windy weather. The surface temperature fell and the water became stirred to a depth of about 17 feet. Below 20 feet, however, there was little change. These

conditions usually continue through the summer, the upper layers becoming warmed and stratified or cooled and mixed, the lower layers remaining stagnant.

On account of the changes of the surface temperature—due to alternations of day and night, sunshine and clouds, winds and calm,—

convection currents are almost continuously at work in the upper strata. Increasing surface temperatures on a sunny day produce a condition of temporary stratification during the day; this is likely to be followed by cooling at night that equalizes the temperatures and mixes the water by vertical convection.

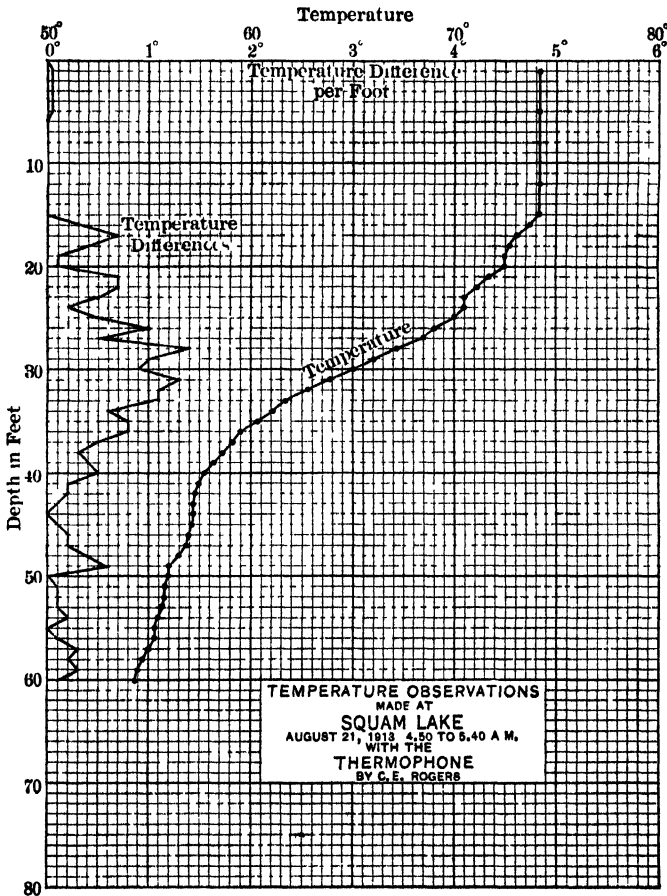


FIG. 43. — Thermocline in Squam Lake. August 21, 1913.

The Transition Zone. — Figures 43 and 44 show the results of temperature observations made at Squam Lake, N. H., during August, 1913, by students taking a course in limnology at the Harvard Engineering Camp.

These diagrams show strikingly that between the upper and lower layers there is a relatively thin stratum where the temperature changes rapidly — sometimes 10° F. in a vertical foot. This region has been

variously named. In Germany it is called the "Sprungschicht"; in Scotland, the "discontinuity layer." Dr. Birge has called it the "thermocline" and has arbitrarily confined its limits to that stratum in which the change in temperature per meter of depth is 1°C or more (0.5°F . per foot). A more satisfactory term, the reasons for which will appear later, seems to be the "transition zone." The position of the transition

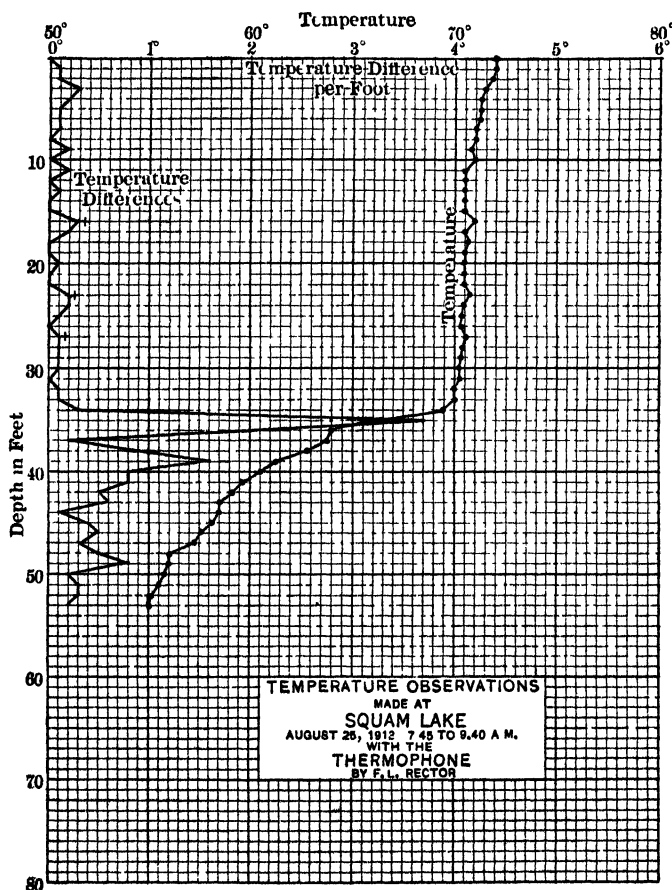


FIG. 44. — Thermocline in Squam Lake August 25, 1913.

zone and the rate of temperature change vary according to the depth of the lake, the intensity of the wind, and the temperature of the water above and below. Its upper boundary is sometimes very sharp, particularly in the autumn; the lower boundary is less distinct. In the fall the transition zone drops toward the bottom as circulation extends to greater and greater depths.

A better conception of the transition zone may be obtained from Fig. 45, which shows the cross-section of a lake as well as the temperature changes. Above the transition zone is the "zone of circulation," and below it, the "zone of stagnation." Dr. Birge calls the region above the transition zone, or thermocline, the "epilimnion," and that below it the "hypolimnion." He has also called the thermocline the "mezolimnion."

Within the circulation zone the water moves horizontally under the influence of the wind, and vertically by convection. It is well aerated and almost decarbonated. In the stagnation zone, however, the horizontal currents are very slight and the vertical currents negligible. The condition of the dissolved gases may then be reversed, oxygen being depleted or even exhausted and carbonic acid increased. Hence the region of rapid temperature change is a transition layer in more ways than one: in temperature, density, viscosity, water movement, and condition of the dissolved gases.

Classification of Lakes According to Temperature.

Lakes may be divided into three *types*, according to their surface temperatures, and into three *orders*, according to their bottom temperatures. The resulting nine classes are shown in Fig. 46. In these diagrams the boundaries

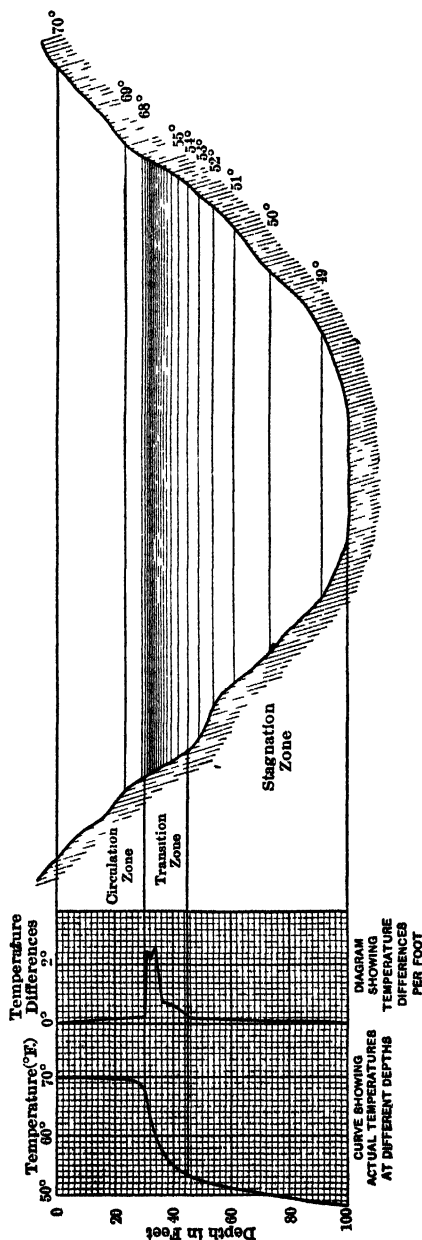


FIG. 45. — Temperature Gradient in a Reservoir and Vertical Zonal Differentiation. (Idealized.)

of the shaded areas represent the limits of the temperature fluctuations at different depths. The horizontal scale represents temperatures in degrees Fahrenheit, increasing toward the right, and the vertical scale represents depths. The three types of lakes are designated as *polar*, *temperate*, and *tropical*. In lakes of the polar type the surface temperature is never above that of maximum density; in lakes of the tropical type it is never below that point; in lakes of the temperate type it is sometimes below and sometimes above it. This division into types corresponds somewhat closely to geographical location.

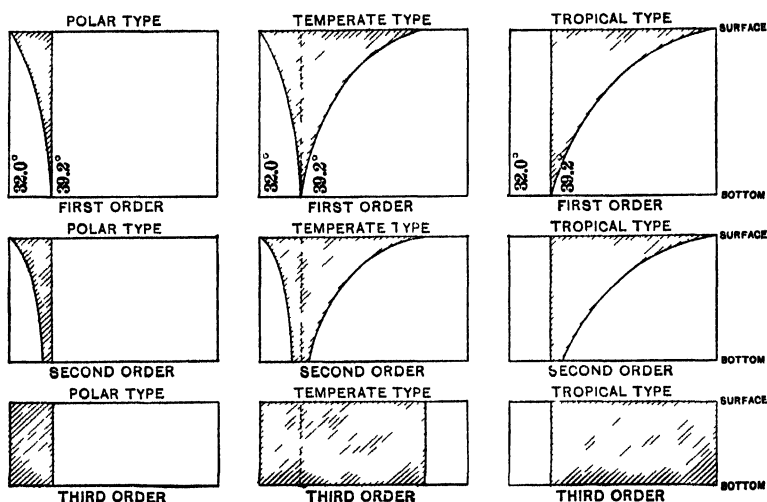


FIG. 46. — Classification of Lakes According to Temperature. Limiting Variations in Vertical Temperature Gradients.

The three orders of lakes may be defined as follows: Lakes of the first order have bottom temperatures that are practically constant at or near the point of maximum density; lakes of the second order have bottom temperatures that undergo annual fluctuations, but are never far from the point of maximum density; lakes of the third order have bottom temperatures that are seldom far from the surface temperatures. The division into orders corresponds in a general way to the hydrography of the lakes; i.e., their size, contour, depth, etc.

The temperature changes that take place in the nine classes of lakes, according to this system of classification, are exhibited in another manner in Fig. 47. The curves in these diagrams trace the surface and bottom temperatures for each season of the year, the dates following the abscissæ, and the temperatures the ordinates. The shaded areas show the difference between the surface and bottom temperatures; the wider the shaded area the greater the difference.

A study of these diagrams brings out some interesting facts concerning the phenomena of circulation and stagnation. In Fig. 46 it is seen that the circulation periods occur when the curve showing the temperatures at various depths becomes a vertical line; that is, when all the water has the same temperature. The stagnation periods are shown by the line being curved, the top to the right when the warmer layers are above the colder, and to the left when the colder layers are above the warmer. In Fig. 47 the circulation periods are indicated by the surface and bottom temperature curves coinciding, and the stagnation

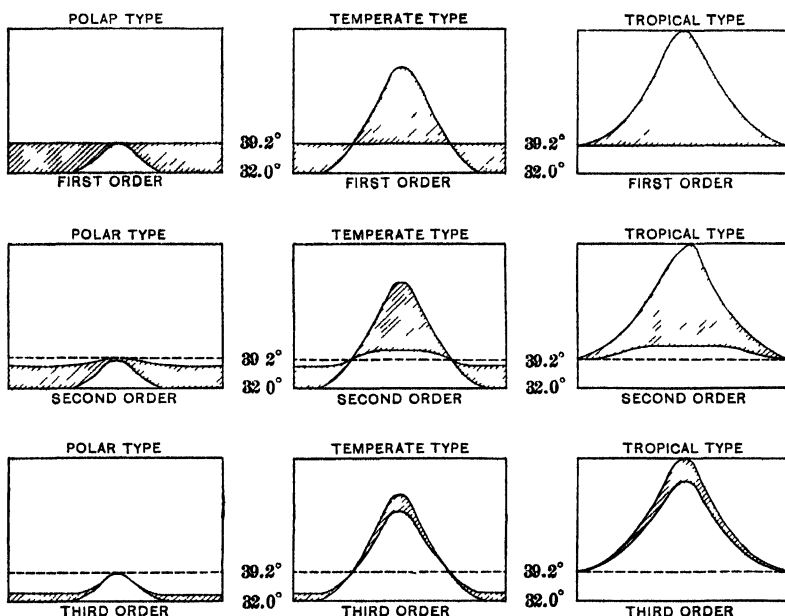


FIG. 47. — Classification of Lakes According to Temperature. Seasonal Variations in Surface and Bottom Temperatures.

periods by their being apart. The distance between the curves indicates, to a certain extent, the difference in density between the top and bottom layers, and we see that the farther apart the lines become the less likelihood there is that the water will be stirred up by the wind.

In lakes of the polar type there is but one opportunity for vertical circulation, except in the third order; namely, in the summer season, when the water approaches the temperature of maximum density. In a lake of the first order, that is, in one where the bottom temperature remains constantly at 39.2° , the circulation period is very short indeed, if not lacking altogether. In a lake of the second order, circulation probably continues for a longer period. In a lake of the third order,

the water is in circulation nearly all the time except when frozen. The minimum temperature limit indicated for this order, i.e., 32° at all depths, is possible only in very shallow bodies of water, and simply indicates that all the water is frozen. The temperature of the ice is most likely below 32° at the surface. It is probable that very few polar lakes exist.

In lakes of the tropical type there is likewise but one period of circulation each year, except in the third order. This does not occur in summer but in winter. In the first order this circulation period is brief or entirely wanting; in the second it is of longer duration; in the third order the water is likely to be in circulation the greater part of the year. Tropical lakes are quite numerous, but observations are lacking to place them in their proper order.

Most of the lakes of the United States belong to the temperate type. In this type there are two periods of circulation and two periods of stagnation except in the third order, as we have seen illustrated in the case of Lake Cochituate. In lakes of the first order the circulation periods are very short or entirely wanting; in the second order the circulation periods are of longer duration; in the third order the water is in circulation throughout the year when the surface is not frozen. The above facts may be recapitulated in tabular form as follows:

TABLE 20
CIRCULATION PERIODS IN DIFFERENT CLASSES OF LAKES

	Polar Type	Temperate Type	Tropical Type
First Order.	One circulation period possible, in summer, but generally none.	Two circulation periods possible, in spring and fall, but generally none.	One circulation period possible, in winter, but generally none.
Second Order.	One circulation period, in summer.	Two circulation periods, in spring and fall.	One circulation period, in winter.
Third Order.	Circulation at all seasons, except when surface is frozen.	Circulation at all seasons, except when surface is frozen.	Circulation at all seasons.

Speaking in very general terms, one may say that lakes of the first order have no circulation; lakes of the third order have no stagnation,

except in winter; and lakes of the second order have both circulation and stagnation.

In view of the comparatively few series of observations of the temperature of our lakes, the author has refrained from making any classification of the lakes of the United States, but the results thus far obtained seem to indicate that the first order includes only those lakes more than about 200 feet in depth, such as the Great Lakes and Lake Champlain; the second order includes those with depths less than about 200 feet, but greater than about 25 feet; and the third includes those with depths less than 25 feet. These boundaries are only approximate, and it should be remembered that depth is not the only factor that influences the bottom temperature.

Stagnation is sometimes observed in small artificial reservoirs even when the depth is less than 20 feet. It is usually of short duration.

WIND ACTION

In discussing the heat conditions of water it has been necessary to explain many of the temperature changes in lakes as due at least in part to wind action. Blowing across the surface of the water, the wind produces waves and currents. Waves are disturbances of the water surface in the form of ridges and troughs. In wave action each particle of water rises, moves forward, sinks, and moves back, describing a circular path that returns it to its original position. Under the influence of wind-induced currents, the surface water drifts across the lake with the wind, but is forced to return when it reaches the windward shore. This it does either by turning aside in the form of eddy currents or by turning down as an undertow. These currents have a very important influence on the lake as an environment of microscopic organisms. The downward-moving surface water on the windward shore carries oxygen to the underlying strata, while conversely the upward-moving bottom water on the leeward shore liberates carbon dioxide at the surface. Foodstuffs of various kinds are moved through the water and reach the layers where organisms abound. The plankton themselves are carried from place to place by the moving water, and the currents flowing over shallow areas are able to pick up spores, or seeds, of organisms, which are thus distributed widely through the lake. This partly explains the rapid seeding of reservoirs. It will be shown later that certain organisms tend to concentrate in the transition zone just below the region of actively circulating water.

Waves. — Waves may be produced in a number of ways but wind waves are by far the most common and the most important. The

characteristics of an oscillatory wave are illustrated in Fig. 48, from which it is seen that each particle of water moves in a circular path which returns it to its original position. In "shallow-water waves," which occur in water of a depth less than half the wave length, the orbits are elliptical instead of circular.

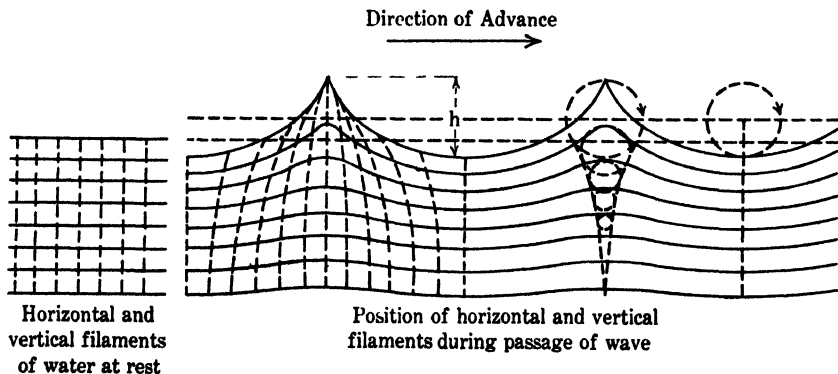


FIG. 48. — Characteristics of Oscillatory Waves.

The height of a wave, namely, the vertical distance from its highest to its lowest point, varies with the clear distance over which the wind can sweep. This distance is known as the "fetch." Hawksley found that the relationship of the wave height, h , in feet produced in large reservoirs by the heaviest gales in England to the length of fetch, f , in feet is $h = .025 \sqrt{f}$.

It follows that the larger a reservoir, and particularly the greater its expanse in the course of the prevailing winds, the more intense will be the wave action. One of the direct results of wave action is the breaking up of filamentous algæ and fragile organisms.

Horizontal Currents. — Although there are some horizontal currents in lakes and reservoirs due to the movement of the water from inlet to outlet, the most important ones are those induced by the wind. The ratio of the velocity of the surface water to that of the air has been shown by experiment to be in the vicinity of 5 per cent in the case of a large lake like Lake Erie. In a small lake it is less than this. Experiments made at Owasco Lake, N. Y., by Ackermann, showed that this ratio decreased as the wind velocity increased, being about 3 per cent for a wind velocity of 5 miles per hour, and about 1 per cent for a wind velocity of 30 miles per hour. Of course, the actual movement of the surface water was greater with the higher wind velocity. In Ackermann's experiments a wind velocity of 5 miles per hour caused the surface water to move at the rate of about 13 feet per minute, while a 30-mile

breeze induced a water movement of 26 feet per minute. Whereas the direction of surface-water movement is generally the same as that of the wind, it is not always so. In small lakes the surrounding topography and the varying contours of the lake bottom influence the direction of the currents.

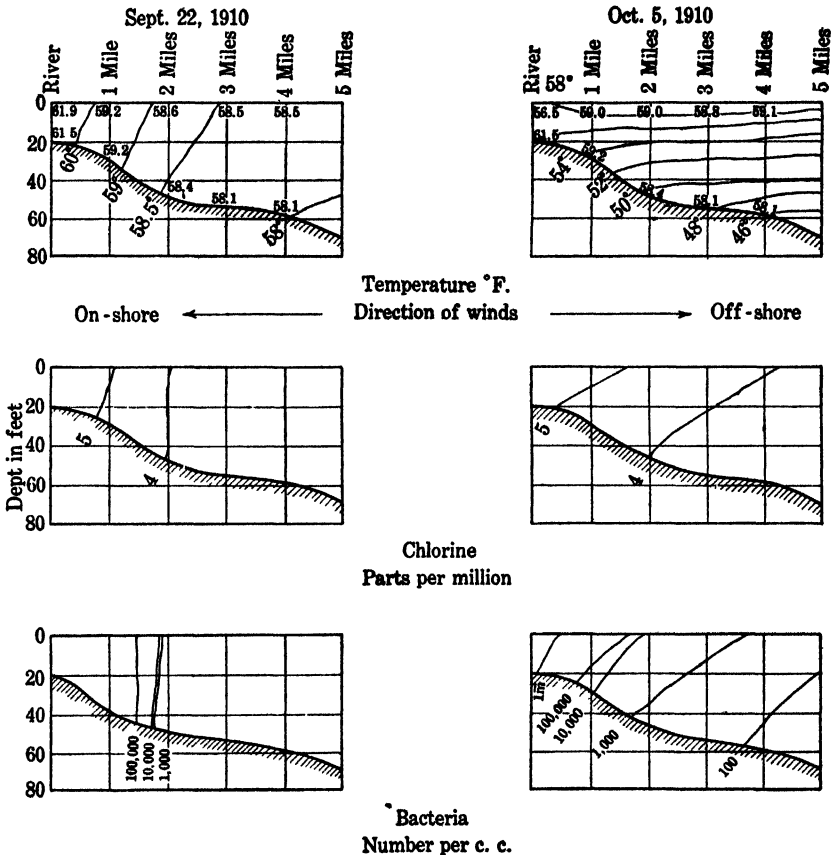


FIG. 49. — Effects of On-shore and Off-shore Winds. Milwaukee Bay, 1910.

The traveling surface water carries the deeper water along with it, but at a slower rate, the velocity decreasing with the depth. At Owasco Lake, the velocity at a depth of 10 feet was about 60 per cent of the surface velocity and decreased to 25 per cent at 20 feet.

Undertow Currents. — As the water in the upper strata is driven toward the windward shore it raises the level there, and the head thus built up produces return currents below the surface. These undertow currents are known to exist at bathing beaches, but it is not so well

of the isotherms. On August 16, for example, the wind was constantly from the southwest, i.e., off-shore, and the isotherms steadily rose as the warmer surface water was carried away. At 4 A.M., August 17, the wind shifted to east and then to northeast, blowing from that direction for nearly 20 hours. Under its influence the warm surface water returned to fill the cove and depress the isotherms.

The nature of the circulation of the water induced by the wind may be finally illustrated by a generalized summary of float experiments made at Squam Lake. Figure 51 shows how floats very near the surface drifted with the wind, while the deeper floats moved in the opposite direction. It was found that the greater part of the return circulation was above the transition zone, but that even below the transition zone there was some movement of the water. When the bottom water is spoken of as stagnant, therefore, it must be understood that this term is not absolutely accurate.

The exact depth of return currents depends upon the temperature

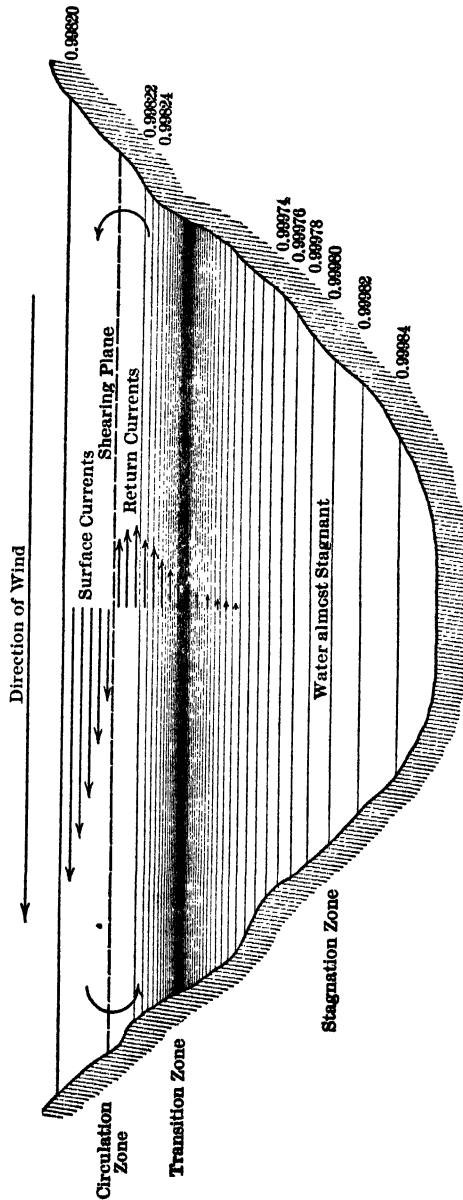


FIG. 51. — Direction and Relative Horizontal Velocity of Wind-induced Currents in a Reservoir Cross-section. (Idealized.) Specific Gravity of the Different Water Strata is shown by Figures at the Right.

of the water. During the periods of complete circulation in the spring and fall, for example, undertow currents may return along the bottom even in very deep lakes. In summer, however, when the lake is vertically stratified, these currents remain largely confined to the circulation zone.

Shearing Plane. — Within the zone of circulation there is a plane which divides the upper currents that follow the wind from the return currents that go counter to the wind. At this plane, called the "shearing plane," a float has almost no motion. The depth of the shearing plane depends to a considerable extent upon the depth of the upper boundary of the transition zone, but it is also influenced by the contours of the lake bottom and by other factors. With increasing wind velocity, more and more water is carried with the surface layers in the direction of the wind, and the stagnant layers are more and more affected by the return currents.

With high winds the upper boundary of the transition zone is more distinct than with light winds.

Seiches. — After a strong wind has been blowing over a lake in one direction for a considerable time and then subsides, the water that has been piled up at the windward end falls to and below its normal level. This is accompanied by a rising of the water at the leeward end of the lake. Then the water on the leeward falls, while that on the windward rises. This synchronous rising and falling of the water gives rise to the phenomenon known as the "seiche" (pronounced *sāsh*). The amplitude of the seiche vibrations may vary all the way from a few hundredths of an inch to several feet, but it is only in very large lakes that large seiches are observed. The time of oscillation is fairly constant for any particular lake. One observer has given the following formula, which, while not accurate, illustrates the nature of the factors involved.

$$t = \frac{2l}{3600 \sqrt{dg}},$$

where t = time of oscillation in hours.

l = length of the lake (or width in the case of transverse seiches) in feet.

d = mean depth in feet of lake along the axis of observation.

g = acceleration of gravity (32.16).

This formula applied to Lake Erie gives a calculated seiche period of 14.4 hours, while the observed periods have ranged from 14 to 16 hours.

Other causes than the wind may produce seiches, such as sudden and unequal changes of barometric pressure at opposite ends of a lake, and

sudden rainfalls at one end of a lake. Seiches are of less importance to the sanitary engineer than are the horizontal currents that accompany them.

Thermal Resistance to Mixture. — The extent to which heat, gases, foodstuffs and organisms are distributed by the wind varies greatly at different seasons of the year. We have seen that it is greatest at the time of the spring and fall overturn when the water can be stirred from top to bottom by relatively light breezes. This variation in behavior of water has been explained by Birge on the basis of thermal resistance to mixture. If we examine the change in density of water for temperature differences of 1°C. , as shown in column (3) of Table 21, we see that it reaches a minimum from 3° to 4° and 4° to 5°C. , i.e., at the temperature of maximum density. It increases thence as the temperature falls or rises. Calling the minimum difference in specific gravity unity, we obtain the relative differences as shown in column (4). From 20° to 21°C. , for example, the relative difference in density is 26.38 times as great as from 4° to 5°C. Since lighter water always overlies denser water, it follows that in order to produce mixture by undertow or other currents the resistance of the water due to differences in specific gravity of the various layers must be overcome by the wind. This resistance is called the "thermal resistance" of the water. As shown above, it becomes greater the farther removed the temperature of the water from that of maximum density.

The fact that thermal resistance to mixture varies in proportion to the difference in densities between the upper and lower strata (assuming a uniform temperature gradient) is obvious. The following mathematical analysis, according to Birge, of the work required to mix a column of water brings out this point.

The work, W , done against gravity in mixing a column of water so that it shall assume uniform density is:

$$W = A \int_0^C f(z) \left[z - \frac{C}{2} \right] dz, \quad (1)$$

where A is the area of cross-section of the column, C the height, and $f(z)$ the function expressing density in terms of z , the distance from the top of the column.

When the temperature gradient is uniform, as assumed in this case, $f(z)$ is a rational integral function of the second degree, since the density, $D = LT^2 + MT + N$, in which T is the temperature and L , M , and N are constants. Equation (1) then reduces to the simple form:

$$W = \frac{AC^2}{12} (D_2 - D_1), \quad (2)$$

TABLE 21
DENSITY OF WATER AND THERMAL RESISTANCE TO MIXTURE
After Birge

Temperature, °C.	Density	Difference in Density for 1° C.	Relative Difference for 1° C.	Work Done in Mixing, Ergs
(1)	(2)	(3)	(4)	(5)
0	0.999868	+0.000059	7.38	0.0491
1	0.999927	+0.000041	5.12	0.0342
2	0.999968	+0.000024	3.00	0.0200
3	0.999992	+0.000008	1.00	0.0067
4	1.000000	-0.000008	1.00	0.0067
5	0.999992	-0.000024	3.00	0.0200
6	0.999968	-0.000039	4.88	0.0325
7	0.999929	-0.000053	6.62	0.0441
8	0.999876	-0.000068	8.50	0.0566
9	0.999808	-0.000081	10.12	0.0675
10	0.999727	-0.000095	11.88	0.0791
11	0.999632	-0.000107	13.38	0.0891
12	0.999525	-0.000121	15.12	0.1008
13	0.999404	-0.000133	16.62	0.1108
14	0.999271	-0.000145	18.12	0.1208
15	0.999126	-0.000156	19.50	0.1299
16	0.998970	-0.000169	21.12	0.1408
17	0.998801	-0.000179	22.38	0.1491
18	0.998622	-0.000190	23.75	0.1583
19	0.998432	-0.000202	25.25	0.1683
20	0.998230	-0.000211	26.38	0.1758
21	0.998019	-0.000222	27.75	0.1849
22	0.997797	-0.000232	29.00	0.1993
23	0.997565	-0.000242	30.25	0.2016
24	0.997323	-0.000252	31.50	0.2099
25	0.997071	-0.000261	32.62	0.2174
26	0.996810	-0.000271	33.88	0.2257
27	0.996539	-0.000280	35.00	0.2332
28	0.996259	-0.000288	36.00	0.2399
29	0.995971	-0.000296	37.25	0.2482
30	0.995673			

where D_1 and D_2 are respectively the density of the upper and lower strata of the column.

When the column of water is 1 meter high and 1 sq. cm. in area and the temperature difference is 1°C. , then W is expressed directly in ergs. Values so figured are shown in column (5) of the table.

From Equation (2) we see that the work required to mix the water is in direct proportion to the difference in density, or, in other words, the thermal resistance to mixture is proportional to the difference in density and therefore varies as the temperature departs from 4°C.

This fact has important and wide application. It explains, for example, why the distribution of heat is so much more rapid in the spring than it is later in the season. The mean temperature in the spring is very close to 4° , and as the mean departs from 4° later in the season the resistance increases, being ten times as great when the temperature has risen to 10° . In other words, the effectiveness of winds of equal intensity is reduced by 90 per cent.

Another fact explained is that the thermocline may be reduced to a temperature amplitude of only 2° in the fall and may thus continue for days or weeks. No such slight difference exists in the spring. The reason is that the temperatures in the spring are in the vicinity of 6° or 8° , while in the fall they lie at 12° or 14° and so offer much more resistance to mixture.

At the junction of the circulation and transition zones the fall in temperature is rapid. A decrease of 4° or 5° per meter is not uncommon, and this is from a high temperature, 20° or more. The thermal resistance to mixture is, therefore, very great, and it is increased by the processes that tend to cause mixture.

In this discussion it has been assumed that water is a perfect fluid. This is not the case; water is viscous and its viscosity is not without influence on the ability of the wind to mix it. Although no quantitative relation between thermal resistance and viscosity has been found, it seems clear that it is not very great. Viscosity increases far more slowly at low temperatures than thermal resistance diminishes.

LIGHT CONDITIONS

Much work must yet be done to explain the behavior of the different species of microorganisms toward light. Most of them, more particularly the chlorophyll-bearing organisms such as the algæ and many of the protozoa, require light for growth and reproduction. They are therefore found in the upper strata of water. The process involved is known as photosynthesis and will be discussed in Chapter VIII. Other organ-

isms shun the light and seek the deeper layers of water where they may utilize only certain radiations of the sun, if any at all. The amount of light received by the microorganisms in a lake depends upon the intensity of the light at the surface of the water and upon the extent to which the light is transmitted by the water. The transparency of water profoundly influences the intensity of light at different depths and hence has a marked effect on the growth of algæ. To compare extreme cases, we observe that when very clear waters, such as ground waters, are exposed to the light in open reservoirs algæ grow abundantly, but that plant life is very meager in the water and along the shores of the silt-laden streams of the Middle West, such as the Mississippi and the Ohio Rivers. Some light is absorbed by all waters, even distilled water, but the amount of light absorbed increases as the suspended matter held by the water increases. As muddy waters become clarified on standing, the growth of organisms tends to increase.

Absorption of Light by Pure Water. — Water, both pure and in its natural state, exhibits the power of selective absorption of light. Because of this fact waters appear colored, the color being determined by the wave length of that radiation of which the greatest percentage is transmitted.

Selective absorption is an electrical resonance phenomenon. Electrons, having definite periods of vibration, give the medium the power to absorb radiations, the frequencies of which agree with those of the electrons. The period depends upon the chemical constitution or arrangement of the atoms in the medium. Atoms existing as elements or in simple inorganic compounds may show an absorption very different from that which they show when they are combined into complicated organic compounds, such as aniline dyes, for which the selective absorption is very great.

The surface of water shows a greater absorption than the deeper layers. Quiescent pure water transmits about 47 per cent of the solar energy through the first meter of depth, 80 per cent of the remaining energy through the 1 to 2-meter stratum, and over 90 per cent through all deeper one-meter strata, the loss per meter rapidly declining to a minimum of about 2 per cent of the energy incident on the upper surface of the stratum. At 5 meters, therefore, there remains about 29 per cent of the original energy of the sun and about 23.4 per cent at 10 meters. When the surface of the water is ruffled by wind action, much of the light incident upon it is reflected and the amount of light transmitted is reduced proportionately. This is also true when the sun's rays strike the water surface at a low angle. The period of daylight under water is therefore much shorter than in the atmosphere.

The absorption is much greater in the first meter of pure water because of the property of selective absorption, i.e., there is a rapid absorption of the rays at the red end of the spectrum as compared with a slow absorption of the shorter waves. This is shown in the following table:

TABLE 22
COEFFICIENTS OF ABSORPTION OF LIGHT
After Pietenpol

Wave Length (Ångström Units)*	Color	Coefficient of Absorption	Wave Length (Ångström Units)	Color	Coefficient of Absorption
4700	blue	.034	6180	orange	.206
4940		.030	6360		.225
5220		.030	6480		.236
5390	green	.022	6630	red	.245
5580		.036			
5790	yellow	.056			
5895		.096			
6005		.165			

* One Ångström unit equals $.0001 \mu = .1 \mu\mu$.

The coefficient of absorption determined by Pietenpol is defined by the equation:

$$e^{cd} = \frac{i}{i'}$$

where e is the base of the Napierian system of logarithms = 2.7183, c is the coefficient of absorption, d the depth of water (1 meter used by Pietenpol), i the intensity of the oncoming light, and i' the resulting intensity.

A coefficient of unity means, therefore, that a stratum of water 1 meter in thickness absorbs so much light that the transmitted light is equal to $\frac{1}{2.7183}$ of its original value. Similarly, an absorption coefficient of 0.200 means that if the ray of light were passed through a layer of water 5 meters thick its intensity would be reduced to $\frac{1}{2.7183}$ of its original value.

Absorption of Light by Natural Waters. — Natural waters exhibit absorptive powers differing from those of pure water and differing be-

tween themselves. This difference may be due either to the stain in the water or to the material giving turbidity to the water.

The effect of turbidity is to increase the absorptive power of the water. The particles do not absorb selectively. This has been shown by Pietenpol. He compared the coefficients of absorption for lake water in its natural condition, after passing through a clarifier and filter, and after having been forced through a Berkefeld filter. The curves given by plotting the coefficient against its corresponding wave length for the three waters were of the same slope, indicating conclusively that the absorption by particles is non-selective in nature, although the coefficient in the case of unfiltered water was approximately twice that of the clarifier effluent and nearly six times that of the Berkefeld filter effluent.

The non-selective property of suspended particles is also shown by the close correlation between transparency and per cent transmission for three New York lakes, as given in Table 23. These lakes have water only slightly stained and differ very little in color, the only varying factor being the turbidity.

TABLE 23
TRANSPARENCY OF LAKE WATER AND TRANSMISSION OF RADIATION
Vertical Sun
After Birge and Juday

	Transparency in Meters	Per Cent Radiations Remaining at 1 Meter	Per Cent Transmission per Meter
Seneca Lake.....	6.8	21.9	72
Cayuga Lake.....	6.2	19.9	66
Canandaigua Lake.....	4.4	19.4	60

The effect of color is to increase the selective absorption of the water. Furthermore, the selectivity varies with the origin of the water, i.e., whether it comes from marshes and swamps or from springs. In the first class, the marsh-stained lakes, the coefficient of absorption rapidly increases when the wave lengths are shorter than 5500 Ångström units. In the case of the second type, the isolated spring-fed lakes, the selective absorption follows more closely that of pure water. Further work on this subject will enable us to classify lakes according to the various

kinds and mixtures of stains that color the water and give rise to different forms of absorption curves.

In general, it may be said that about 71 per cent of the solar energy received is transmitted through each meter stratum in the case of natural waters. At this rate only 5.4 per cent of the original sun's energy will be found at a depth of 5 meters, and 1 per cent at 10 meters, instead of 29 per cent and 23.4 per cent as in the case of pure water. The wide difference is due to the suspended matter which possesses large non-selective absorptive powers. The results of Birge and Juday's study of the Finger Lakes of New York are given in Table 24:

TABLE 24
TRANSMISSION OF SUN'S ENERGY IN LAKE WATER
After Birge and Juday

Per cent of the energy found at the upper surface of each 1-m. stratum which is present at the lower surface of that stratum.

Stratum, in Meters	Transmission, Per Cent
0-1	21.4
1-2	70.2
2-3	72.6
3-4	71.0
4-5	69.6
5-6	69.8
6-7	71.1
7-8	67.8
8-9	72.2
9-10	75.9

In connection with the Finger Lake studies, the electrical pyrlim-nometer was successfully used. This instrument consists of a receiver containing 20 small thermal couples. It is lowered into the lake to any desired depth and alternately exposed to the sun and covered. The electrical effect of the sun's radiation on the thermal couples is proportional to the energy in its rays, and the resulting electrical currents are measured by the deflection caused in a d'Arsonval galvanometer.

Shelford and Gail used a potassium hydroxide photo-electric cell in their absorption studies at the Puget Sound Biological Station. From the results of their studies they concluded that about 25 per cent of the light is shut out by the surface of the water and 20 per cent by the first

meter. They found 8 to 10 per cent of the shorter wave lengths entering the surface at a depth of 10 meters.

The reduction of light in passing downward through a body of water follows in general the law that as the depth increases arithmetically the light decreases geometrically. This is borne out by data given by Birge and Juday for Seneca Lake. The relation in this case may be expressed by the equation:

$$i = 0.43 e^{-0.00346d},$$

where i is the intensity of light as measured by the electrical pyrlimnometer, expressed in calories per square centimeter per minute, and d is the depth in centimeters.

Transparency of Lakes. — A simple estimate of the transparency of a certain body of water may be had by using the methods originally employed by Forel and others in Switzerland. Three methods of experiment were employed. The first was that of the visibility of plates. This method, used by Secchi in 1865 in determining the transparency of the water in the Mediterranean Sea, consisted of lowering a white disk (20 cm. in diameter) into the water and noting the depth at which it disappeared from view, and then raising it and noting the point at which it reappeared. The mean of these two depths was called the limit of visibility. The second method, known as that of the Genevan Commission, was similar to the first, but instead of a white disk an incandescent lamp was lowered into the water. This light when seen through the water from above presented an appearance similar to that of a street lamp in a fog; that is, there was a bright spot surrounded by a halo of diffused light. When the light was lowered into the water the bright spot first disappeared from view. The depth of this point was noted as the "limit of clear vision." Finally the diffused light disappeared and the depth of this point was called the "limit of diffused light." Both these methods were useful only in comparing the relative transparency of different waters or of the same water at different times. In order to get an idea of the intensity of light at different depths a photographic method was used. Sheets of sensitized albumen paper were mounted in a frame in such a way that half of the sheet was covered with a black screen, while the other half was exposed. A series of these papers was attached to a rope and lowered into the water; they were equidistant and so supported that they assumed a horizontal position in the water. They were placed in position in the night and allowed to remain 24 hours. On the next night they were drawn up and placed in a toning-bath. A comparison of prints made at different depths enabled the observer to determine the depth at which the light ceased

to affect the paper and to obtain an idea of the relative intensity of the light at different depths. To assist in this comparison an arbitrary scale was made by exposing sheets of the same paper to bright sunlight for different lengths of time.

The results of the experiments are given by Forel as follows:

In Lake Geneva the limit of visibility of a white disk 20 cm. in diameter was 21 m. The limit of clear vision of a 7-candlepower incandescent lamp was 40 m.; the limit of diffused light was about 90 m. The depth at which the light ceased to affect the photographic paper was 100 m. when the paper was sensitized with chloride of silver, and about 200 m. when sensitized with iodobromide of silver. These depths were less in summer than in winter on account of the increased turbidity of the water. The transparency of the water in other lakes, as shown by the limit of visibility of a white disk, is cited as follows: Lake Tahoe, 33 m.;

La Mer des Antilles, 50 m.; Lac Lucal, 60 m.; Mediterranean Sea, 42.5 m.; Pacific Ocean, 59 m. It should be remembered that these are all comparatively clear and light-colored water, and that in them the light penetrates to far greater depths than in turbid and colored water. For example, in Chestnut Hill reservoir, a disk lowered into the water at a time when the color was 92 p.p.m. disappeared from view at a depth of six feet.

The author's experiments have shown that the limit of visibility may be determined most accurately by using a disk about 8 inches in diameter, divided into quadrants painted alternately black and white like the target of a level-rod, and looking vertically down upon it through a water telescope provided with a suitable sunshade (Fig. 52). It has been found that the limit of visibility obtained in this manner bears a close relation to the turbidity of the water as determined by a turbidimeter. It also varies with the color of the water, but the relation has not been carefully worked out.



FIG. 52.—Disk for Comparing the Transparencies of the Water in Different Lakes.

Color of Water. — Some surface waters are colorless, but in most ponds and lakes the water has a more or less pronounced brownish color. This may be so slight as to be hardly perceptible, or it may be as dark as that of weak tea. It is darkest in water draining from swamps, and the color of the water in any pond or stream bears a close

relation to the amount of swamp land upon the tributary watershed. The surface water in granite regions is generally darker than in regions of shale or slate.

The color is due to dissolved or colloidal substances of vegetable origin extracted from leaves, peaty matter, etc. It is quite as harmless as tea. The exact chemical nature of the coloring matter is not known. It is complex in composition. Tannins, glucosides, and their derivatives are doubtless present. The color of water usually bears a close relation to the albuminoid ammonia present. Carbon, however, is the important element in its composition. Hence the color of water also varies closely with the "oxygen consumed." Iron is present, and its amount varies with the depth of the color. In some waters iron alone imparts a high color, but in peaty waters it plays a subsidiary part. Manganese may also play a part.

The color of a water is usually stated in figures based on comparisons made with some arbitrary standard, the figures increasing with the depth of the color. The Platinum Cobalt Standard, the Natural

Water Standard, and the Nessler Standard are those which have been most commonly used. The first is now the accepted standard.*

Determination of Color in the Field. — For field work a color comparator, by which the color of the water is compared with that of disks of colored glass, is very useful. The water is placed in a metallic tube with glass ends. Its color is then compared with the color produced by covering one end of a second tube containing distilled water with one or

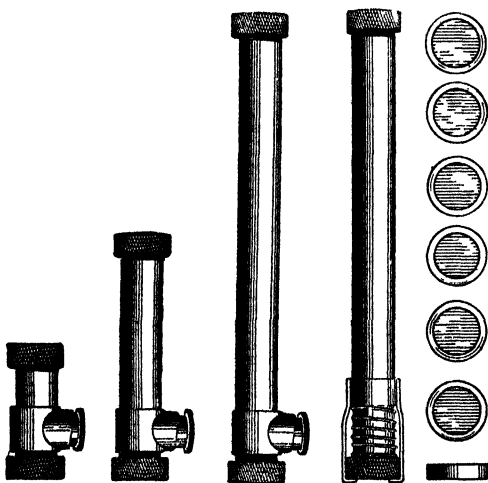


FIG. 53. — U. S. Geological Survey Field Apparatus for Measuring the Color of Water.

more of the glass disks. This apparatus, devised for the United States Geological Survey by Dr. Allen Hazen and the author, is illustrated in Fig. 53.

The amount of color in the water collected from a watershed is subject to seasonal variation. This may be illustrated by the color of the water in Cold Spring Brook, at the head of the Ashland Reservoir of the

* See Standard Methods of Water Analysis.

Boston Metropolitan Supply. This brook is fed in part from several large swamps. The figures given are based on weekly observations.

TABLE 25
AVERAGE COLOR OF WATER IN COLD SPRING BROOK. 1894
Results are Expressed as Parts per Million of Platinum

January..... 99	April..... 93	July.. 98	October..... 69
February..... 88	May.....142	August... .. 75	November....144
March..... 96	June.....159	September... 60	December....120
			Average.....104

There are usually two well-defined maxima, one in May or June and one in November or December. In the winter and early spring the color of the water is low because of dilution by the melted snow. As the yield of the watershed diminishes the color increases until the water standing in the swamp areas ceases to be discharged into the stream. During the summer the water in the swamps is high-colored, but its effect is not felt in the stream until the swamps overflow in the fall. Heavy rains during the summer may cause the swamps to discharge and increase the color of the water in the reservoirs below. It has been found that in general the color of the water delivered from any watershed bears a close relation to the rainfall. In some localities this is more noticeable than in others. In Massapequa Pond of the Brooklyn water supply the color varies greatly from week to week, and the fluctuations are almost exactly proportional to the rainfall. In large bodies of water the seasonal fluctuations in color are less pronounced.

The hue of the water in the autumn is somewhat different from that in the spring. The fresh-fallen leaves and vegetable matter give a greenish-brown color that is quite different from the reddish-brown color produced from old peat.

Bleaching of Colored Water. — When colored water is exposed to the light it bleaches. A series of experiments made at the Boston Water Works by exposing bottles of high-colored water to direct sunlight for known periods showed that during 100 hours of bright sunlight the color was reduced about 20 per cent, and that with sufficient exposure all the color might be removed. The bleaching action was found to be independent of temperature. Sedimentation had but little influence on it. It was dependent entirely upon the amount of sunlight. The percentage reduction was independent of the original color of the water.

This bleaching action takes place in reservoirs where colored water is stored. Stearns has stated that in an unused reservoir 20 feet deep the

color of the water decreased from 40 to 10 p.p.m. in six months. In the Ashland reservoir referred to, the average color of the water in the influent stream for the year 1894 was 104. For the same year the average color of the water at the lower end of the basin was 71. It should be stated that this difference is not due wholly to bleaching. The amount of coloring matter entering the reservoir is not correctly shown by the figure 104, for the reason that the quantity of water flowing in the stream is not uniform. It is greatest in the spring when the melting snows give the water a color lower than the average. Furthermore, a certain amount of colorless rain water and ground water enters the basin. There is also a loss of high-colored water at the wasteway at a season when the color of the water is above the average. It is a difficult matter to ascertain the exact amount of bleaching that takes place in a reservoir through which water is constantly flowing.

Experiments (by the author) made by exposing bottles of colored water at various depths in reservoirs have shown that the bleaching action that takes place at the surface of a reservoir is considerable, sometimes 50 per cent in a month. It decreases rapidly with increasing depth, and the rapidity with which it decreases below the surface depends upon the color of the water in the reservoir, as the following table will show:

TABLE 26
EXPERIMENTS TO DETERMINE THE AMOUNT OF BLEACHING ACTION AT DIFFERENT DEPTHS

	Expt. No. 1	Expt. No. 2	Expt. No. 3
Color of water in reservoir. . . .	20 p.p.m.	37 p.p.m.	44 p.p.m.
Time of exposure.	Aug. 6-Sept. 4	May 5-June 4	July 2-Aug. 3
Color of water exposed.	175 p.p.m.	272 p.p.m.	170 p.p.m.
Percentage reduction of color:			
At depth of 0.0 ft.	52	41
" " " 0.5 "	65	29	20
" " " 1.25 "	32	8	12
" " " 2.5 "	21	4	4
" " " 5.0 "	14	4	3
" " " 7.5 "	3	0	0
" " " 10.0 "	1	0	0
" " " 15.0 "	0	0	0
Dark room.	0	0	0

From these and many similar experiments it has been found possible to calculate the extent of the bleaching action that takes place in any reservoir. The results agree closely with the observed color readings

of the water in the reservoir. The experiments also bear directly upon the penetration of light into the water of a reservoir.

Turbidity of Water. — The turbidity of water is due to the presence of particles of matter in suspension, such as clay, silt, finely divided organic matter, and microscopic organisms.

Three principal methods are used for measuring turbidity. They are: 1, Comparison with silica standards. 2, Platinum-wire method. 3, Turbidimeter method. In all cases the results of the observations are expressed in numbers that correspond to turbidities produced by equivalent parts per million of finely-divided silica. The three methods give fairly comparable results.

The standard of turbidity has been defined by the U. S. Geological Survey as follows:

“The standard of turbidity shall be a water which contains 100 parts of silica per million in such a state of fineness that a bright platinum wire 1 millimeter in diameter can just be seen when the center of the wire is 100 millimeters below the surface of the water and the eye of the observer is 1.2 meters above the wire, the observation being made in the middle of the day, in the open air, but not in sunlight, and in a vessel so large that the sides do not shut out the light so as to influence the results. The turbidity of such water shall be 100.”

Determination of Turbidity in the Field. — The most convenient method for limnological field-work is the platinum-wire method. This method requires a rod with platinum wire of a diameter of 1 mm. or 0.04 inch, inserted in it about one inch from the end of the rod and projecting from it at least one inch at a right angle. At a distance of 1.2 meters (about 4 feet) above the wire is placed a mark which indicates the position of the observer's eye. The rod is graduated as follows:

The graduation mark of 100 is placed on the rod at a distance of 100 mm. from the center of the wire. Other graduations are made according to Table 27 which is based on the best obtainable data and in which the distances are intended to be such that when the water is diluted the turbidity reading will decrease in the same proportion as the percentage of the original water in the mixture. These graduations are those used to construct what is known as the U. S. Geological Survey Turbidity Rod of 1902. (See Fig. 54.)

Procedure. — “Push the rod vertically down into the water as far as the wire can be seen, and then read the level of the surface of the water on the graduated scale. This will indicate the turbidity.”

The following precautions should be taken to insure correct results:

“Observations should be made in the open air, preferably during the middle of the day and not in direct sunlight. The wire should be

kept bright and clean. If for any reason observations cannot be made

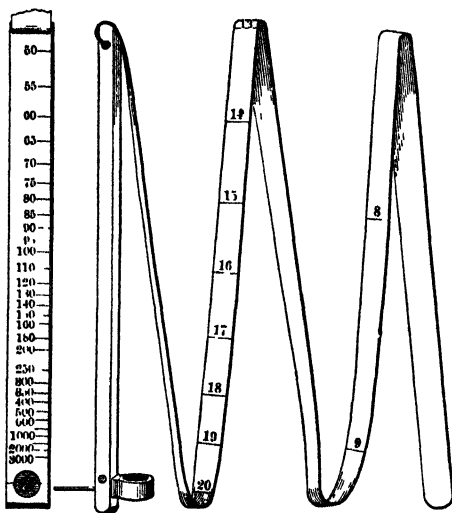


FIG. 54. — U. S. Geological Survey Turbidity Rod.

directly under natural conditions a pail or tank may be filled with water and the observation taken in the receptacle. In this case the water should be thoroughly stirred before the observation is made, and the diameter of the vessel should be at least twice as great as the depth to which the wire is immersed. Waters that have a turbidity above 500 p.p.m. should be diluted with clear water, and the degree of dilution used should be stated and form a part of the report."

For very clear waters the use of a black-and-white disk, as already suggested, will be found

more satisfactory than the platinum-wire method.

TABLE 27
GRADUATION OF TURBIDITY ROD

Turbidity, Parts per Million	Vanishing Depth of Wire, mm.	Turbidity, Parts per Million	Vanishing Depth of Wire, mm.	Turbidity, Parts per Million	Vanishing Depth of Wire, mm.
7	1095	28	314	120	86
8	971	30	296	130	81
9	873	35	257	140	76
10	794	40	228	150	72
11	729	45	205	160	68.7
12	674	50	187	180	62.4
13	627	55	171	200	57.4
14	587	60	158	250	49.1
15	551	65	147	300	43.2
16	520	70	138	350	38.8
17	493	75	130	400	35.4
18	468	80	122	500	30.9
19	446	85	116	600	27.7
20	426	90	110	800	23.4
22	391	95	105	1000	20.9
24	361	100	100	1500	17.1
26	336	110	93	2000	14.8
				3000	12.1

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CHAPTER VIII

LIMNOLOGY — CHEMICAL CONDITIONS

Consideration of the chemical aspects of limnology involves a study of many different compounds and classes of substances. Inasmuch as these influence the fertility of lakes and ponds with respect to the development of plankton life the chemistry of such waters is important in limnological studies. This chapter will deal with the chemical conditions that affect the growth of microscopic organisms in lakes and ponds, the origin and utilization of substances that serve as food and certain phenomena that appear in the chemical arena.

No form of life will survive or perpetuate itself in chemically pure water. Food is a fundamental requirement of the living cell; hydrogen and oxygen alone do not comprise all the elements that enter into the composition of cell material. Natural waters, however, are never chemically pure, for water, in its rôle of the most nearly universal solvent known, dissolves gases from the air and soil, mineral salts from the earth's crust, and organic matter from plant and animal refuse. In addition, the marked erosive powers of water cause considerable amounts of insoluble mineral and organic substances to be washed into streams and lakes in suspended form. The variety of these impurities is wide enough to embrace all the necessary food elements for aquatic forms of life.

MAGNITUDE OF AQUATIC GROWTHS

When coupled with proper environmental conditions the supply of nourishment in natural bodies of water produces growths of plant and animal life which, even though they be made up of microscopic individuals, rival in bulk the crops put forth by the soil. The analogy between the yield of the aquatic environment and that of the land has been so well pictured by Kofoid that his description is here quoted at length.

The land and water share alike the capacity of production of an annual crop of vegetation and of animal life feeding thereon, dependent upon the seasonal course of solar illumination. Thus the soil produces its annual crop of grass, the trees their annual crop of leaves and wood, and these in turn support an insect fauna, and under man's control the fields have an annual pasturage value which can be stated in pounds per acre of beef, pork, or mutton. We may also state the result in chemical terms of the amounts of cellulose, starch, sugar, and protein which the vegetable world produces and assess its food values in terms of calories. In the same way the per-

manent bodies of fresh water, including reservoirs, which man constructs, also produce their annual tonnage of vegetable growths and, if stocked, their annual returns in fish, while the sea produces its seasonal crop of oysters, lobsters, shrimps, fish, and whales which directly or indirectly gain their support from the plant life of the ocean.

This plant life of water, however, differs from that of the land in not building up large growths, such as the grasses, cereals, alfalfa and trees, but in being composed almost wholly of low and simple, primitive forms of vegetable life known as diatoms, desmids, dinoflagellates and algæ. These are photosynthetic organisms which multiply with astounding rapidity and produce throughout the year successive crops of microscopic vegetation. These are the primitive food supply of the animal world of the water, which in turn is in part itself microscopic and fluctuates in quick response to the ever-changing quantities of plant life. These quantities of the plant life are produced in rapidly succeeding waves or pulses of production throughout the year and owe their changing dimensions to the fluctuations in light as influenced by the length of the day, the amount of sunlight and primarily the total amount of radiant solar energy which reaches the water. The time factor, the chemical substances in solution, and the temperature modify the amount and the rate at which growth proceeds and thus determine the tonnage per acre of the reservoir.

Quantitative measurements and chemical analysis of production per acre of water surface during the year indicate that the production in the water does not differ greatly from that of the adjacent land in the total amount of organic matter or available food supply which is produced. It is, however, spread over more of the year, is not accumulated in a single harvest time, but recurs over and over again throughout much of the whole year in minor harvests which represent a fraction of the year's production. Chemical analysis of the plankton of the sea or of fresh water differs but little from that of the rye crop, or of alfalfa. The tonnage of fish produced per acre of a reservoir in the course of a year would approximate that of the normal tonnage of beef produced by the meadows on an acre of land.

The accompanying table of analyses is reproduced from Brandt's investigation of the food values of autumn plankton of fresh water, good meadow hay and of lupines on the one hand, and of Ceratium plankton, poor meadow hay and of green rye on the other. These reveal a rather close equivalence of values between fodders of the land and the products of the microscopic water meadows as measured in albumens, fats, carbohydrates, N-free extract and raw fiber.

TABLE 28
COMPARATIVE FOOD VALUES OF LAND AND WATER CROPS

	Percentage Composition					
	Albumen	Fat	Carbo- hydrates	N-Free Extract	Raw Fiber	Ash
Rich meadow grass.....	20.6	4.5	64.6	10.1
Autumn plankton.....	20.2-21.8	2.1-3.2	60-68.9	8.5-15.7
Lupine.....	20.6	2.6	72.0	4.6
Poor meadow hay.....	8.7	1.7	44.5	39.1	5.8
Rye straw.....	3.5	1.5	38.8	51.3	4.7
Peridinids, Ceratium.....	13.0	1.3	39.0	41.5	5.2
Good hay.....	13.6	3.2	48.2	26.8	8.2
Green rye.....	12.0	3.3	51.6	27.0	5.8

The seasonal sequences of the changes of the vegetation and the accompanying animal life in the reservoir are not unlike those of the land in certain striking particulars. In the first place periods of rapid growth occur at the same seasons of the year in both regions. When the elms and maples are leafing out in early spring in April on the shore of the reservoir, the water itself is flushed with the fresh growth of diatoms and algæ. These growths occur year after year with the same regularity as does the bursting of the buds of the maple or the catkins of the willow, and are exquisitely timed to certain sum totals of heat which may be measured by adding up the total heat of each day as the season progresses. In some years the total heat attains the flushing level earlier than others and we have an early spring both on land and in the reservoirs.

The autumn season on land often brings a recurrence of a second and minor period of renewed growth of vegetation, and reservoirs repeat this with an autumnal flush of growth of diatoms, similar to that of the spring, but of lesser volume. The winter is a season of lessened production, but of different species as a whole from that of the summer, and the summer months a season of recurrent minor flushes not unlike the successive crops of alfalfa that may be reaped from the meadow lands. These, however, do not equal in volume the production of the spring and fall as a general rule.

VITAL PROCESSES AND DISSOLVED GASES

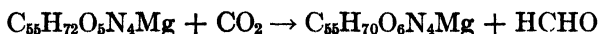
There are present in water three manifestations of the life processes of microscopic cells that deserve mention because of the way in which they affect the content of dissolved gases and, therefore, the food supply of the plankton. These are photosynthesis, respiration, and bacterial decomposition.

Photosynthesis. — Most of the microscopic organisms that are of interest to the water supply specialist belong to, or are closely related to, the plant kingdom and within their cells contain chlorophyll. By virtue of this substance they have the synthetic power of food building by which carbon dioxide and water are united to form formaldehyde, starch and other carbohydrates. The process, which takes place only in the presence of light, is known as photosynthesis.

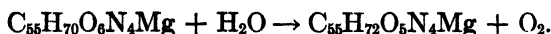
Nature of Photosynthesis. — Although the exact nature of photosynthetic activity is not well understood, there are certain fundamental facts that are recognized. It is well known, for instance, that such external factors as light intensity, temperature and partial pressure of carbon dioxide exercise important influences. There is, also, a general belief that internal factors are operative, although in a way and to an extent not fully appreciated. These include the relation of the chlorophyll pigment to photosynthetic activity, dependence of photosynthesis upon oxygen pressure, and the effect of toxins and other poisons upon the reactions. The close interrelation of all these factors, so common in biological phenomena, makes difficult a true estimate of the importance of each.

*

The first chemical change in the process is an interaction between carbon dioxide and water under the influence of light and acting through chlorophyll. This leads to the formation of formaldehyde, which at once undergoes a condensation, forming sugars, starches and celluloses. It is probable that chlorophyll enters into the reaction, uniting with carbon dioxide to produce chlorophyll B and formaldehyde, as follows:



and that water may then unite with chlorophyll B to produce chlorophyll A and to liberate oxygen.



Light is necessary for both these reactions. The assumption that there is a continual reconstruction of the chlorophyll molecule is borne out by the work of Dixon, Ball, Ewart and others.

Spectrum Influences. — Recent investigation seems to point to the fact that the organisms are more sensitive to the blue end of the spectrum. This fact, that a greater sensitiveness is exhibited toward a certain wave length of light, is in accord with the electronic theory. The work of Pietenpol on selective absorption in the visible spectrum by different lake waters is of importance in this connection, the varying absorption having a direct bearing upon the nature of the planktic life found in the lakes. A few simple experiments on the effect of ultra-violet light and sunlight upon algæ, carried out by T. F. Hatch in the Department of Sanitary Engineering at Harvard University, are of interest in this connection. The samples were taken from Fresh Pond, Cambridge, and contained 1484 standard units of microscopic organisms per cc. The microscopical counts in the various samples after exposure are given in the following table.

It will be noticed that the sample kept in the dark changed only slightly from the original sample. Both sunlight and ultra-violet light seem to have caused a devitalizing of the diatomaceæ, excepting *Synedra*, with a stimulation of the chlorophyceæ and the cyanophyceæ, the ultra-violet light providing a slightly greater stimulus.

One reason why photosynthesis is so little understood is because of the difficulties of experimentation. Gail has made use of the oxygen formed by the reaction as a measure of relative activity. In his work he placed approximately equal amounts of algal growth in each of two bottles, one being of clear glass and the other blackened to exclude light. The water in the bottles contained a known amount of dissolved oxygen. By exposing the two bottles at the desired location, the resulting activity could then be measured. The increase in dissolved

oxygen in the clear bottle was the resultant of photosynthetic activity and of respiration. The depletion in the blackened bottle was due to respiration. The sum of the two was the amount of oxygen produced by photosynthesis and could be compared directly with that formed by the same amount of material in water containing the same initial oxygen content but exposed under different conditions. By securing several pairs of bottles at intervals on a cable they may be lowered into a lake and the intensity of light in relation to depth studied.

TABLE 29
EFFECT OF SUNLIGHT AND ULTRA-VIOLET LIGHT ON PLANKTON GROWTH

Species	Standard Units per cc.			
	Original Sample	After 50 Hrs. of Sunlight	After 50 Hrs. of Ultra-Violet Light	Kept in Dark
Asterionella.....	764	Disintegrated	...	400
Melosira.....	188	60	52	210
Stephanodiscus.....	232	Disintegrated	16	230
Synedra.....	36	74	204	60
Tabellaria.....	132	18	...	26
Pandorina.....	...	88	92	...
Scenedesmus.....	...	56
Staurastrum.....	20	62	154	36
Microcystis.....	...	114	110	...
Peridinium.....	64	...	12	...
Trachelomonas.....	...	36	28	78
Actinophrys.....	48
Total.....	1484	508	568	1040

Photosynthesis in lakes is confined by reason of light requirements to the strata relatively near the surface. In turbid water it is limited to depths of a few inches; in very clear water it may take place at depths of 25 feet or more, although at these depths its activity is slight. In photosynthesis carbon dioxide is taken in by the growing vegetation while oxygen is given off.

Respiration. — Another manifestation of the life processes of microscopic cells is summed up in the word respiration, which is common to both animals and plants. By it oxygen is taken in, and carbonic acid

given off, the released energy appearing as heat and work in the cells. Unlike photosynthesis the process of respiration goes on in the dark as well as in the light. In the light, however, the respiration of green plants may be masked so far as gas relations are concerned by the greater effects of photosynthesis. Animal organisms, such as the holo-zoic protozoa, rotifera and crustacea, and the fungi, which contain no chlorophyll, do not have the photosynthetic power of food building and hence must consume organic food that has already been prepared.

Bacterial Decomposition. — The bacterial organisms are, as a class, not included in microscopical studies of water. Their activities are, however, important in the relation they bear to the food supply of the plankton. Bacteria in water live upon dead organic matter, taking in oxygen and giving off carbon dioxide. Their activity exercises a strong controlling influence upon the amount of these gases that is present at any time. If slight, the depletion of oxygen and the increase in carbon dioxide is masked by the normal interchange of gases between water and air; if pronounced, there is a disturbance of normal conditions with a marked increase in the amount of carbon dioxide, and a depletion in dissolved oxygen that may result in its total absence. In the latter case the bacteria must then derive oxygen from the organic food itself. The resulting decomposition gives rise not only to carbon dioxide but, also, to carbon monoxide, methane and other partially oxidized products of organic matter. This process has sometimes been called anaërobic respiration, — that is respiration without air. It is also known as putrefaction. Bacterial decomposition under anaërobic conditions takes place at the bottom of deep lakes where the water lies stagnant for long periods because of thermal stratification. In these stagnant layers there is no opportunity for exchange of gases with the air; there is always a tendency for oxygen to become depleted and for carbon dioxide to increase in amount.

PLANKTON REQUIREMENTS

Food. — The kind and abundance of food are the most important controlling influences in the life of the plankton. Fluctuations in the food supply, either qualitative or quantitative, affect growth and reproduction and largely determine the onset and number of recurrent pulses of growth. The phytoplankton, made up of plant forms of microscopic organisms, primarily requires water, carbon dioxide, oxygen, nitrates and phosphates. All of these inorganic substances are found to some extent in the waters of lakes, ponds and reservoirs. Other common salts and compounds such as the bicarbonates, chlorides and

sulphates of sodium, potassium, calcium, magnesium, and iron, together with silica, alumina and manganese compounds may exercise a stimulating effect upon certain species or be drawn upon when substances more easily utilized are deficient in amount. There is also evidence that points to the absorption, by some species, of organic pabulum, such as amino-acids, fatty acids, skatol, urea, and humus acids.

The true animal forms of microscopic organisms, which make up the zoöplankton, cannot as a class subsist upon inorganic food because of differing metabolism. Their requirements are principally met by consumption of the phytoplankton and bacteria, also, by utilization of other organic materials. In relation to aquatic life as a whole the plant forms of the plankton are food producers, or synthetic organisms, the animal forms food consumers, or analytic organisms.

Chemical Analysis of Microörganisms.—The utilization of various chemical elements by the growing cells of organisms, which implies a need for these elements, may be studied by chemical analyses of mass growths of different genera and species. Considerable knowledge regarding necessary food has been acquired by this procedure. The chief shortcoming of such studies is that the analysis of cell material deals with living cell substance, reserve food substances, and the waste products of cell activity, and does not indicate the form in which the various elements can be assimilated for nutritional purposes. Tables 30 to 32 present the results of some exhaustive experiments to determine the composition of typical plankton organisms.

In Part II of Table 30 the values for various organic constituents are those given in Part I after recalculation to include the percentage of ash found. Distribution of the ash content was in proportion to the percentage of the various organic compounds as reported in Part I. This was done to avoid the impression that the ash content exists by itself in the cell apart from the complex organic structure.

The cyanophyceæ when compared with other organisms are high in nitrogen and, therefore, in protein. Evidence of this is often given by the strong, disgusting odors of decay that result from the death and decomposition of large growths of cyanophyceæ. The complex protein molecule produces sulphur and nitrogen compounds with strong physical properties. Diatomaceæ and certain chlorophyceæ contain only half as much protein as the cyanophyceæ. Among animal forms the crustacea are high in protein. They are also very high in ether-soluble matter, as are the diatomaceæ.

The large content of pentosans, or gums, ascribed to *Spirogyra* and *Cladophora* in Table 30 marks a characteristic different from other

forms. Conspicuous also is the large amount of crude fiber in *Cladophora* and in the *Peridiniidæ*.

TABLE 30
CHEMICAL ANALYSES OF VARIOUS ORGANISMS
Stated in Percentages of the Dry Weight of the Sample
After Birge and Juday

PART I. ASH INCLUDED

Organisms	Nitrogen	Crude Protein (N×6.25)	Ether Extract	Pentosans	Crude Fiber	Nitrogen Free Extract	Ash	SiO ₂
<i>Cyanophyceæ</i> :								
<i>Microcystis</i>	8.60	53.75	4.55	...	2.11	32.05	7.55	0.38
<i>Microcystis</i>	9.25	57.94	2.67	4.97	0.26	34.82	4.31	0.13
<i>Microcystis</i>	6.32	39.50	2.75	...	0.65	52.09	5.01	...
Chiefly <i>Microcystis</i>	8.35	52.19	5.02	7.80	7.81	1.62
<i>Anabæna</i>	8.27	51.69	1.11	4.81	0.63	39.40	7.17	0.95
<i>Anabæna</i> and <i>Cœlo-</i> <i>sphaerium</i>	8.35	52.19	2.05	6.15	1.17	39.93	4.66	0.27
<i>Aphanisomenon</i>	9.30	58.12	3.72	2.04	0.53	30.12	7.51	1.16
<i>Aphanisomenon</i> and <i>Anabæna</i>	9.94	62.12	4.34	3.42	1.30	25.72	6.52	0.17
<i>Lyngbya</i>	9.17	57.31	2.36	5.25	3.42	31.24	5.07	0.15
<i>Lyngbya</i>	8.21	51.31	1.38	3.76	7.39	34.74	5.18	0.20
<i>Diatomaceæ</i>	3.66	22.87	13.60	2.87	1.43	22.60	39.50	30.78
<i>Chlorophyceæ</i> :								
<i>Volvox</i>	7.61	47.56	5.54	1.00	6.32	34.30	6.28	0.24
<i>Spirogyra</i>	3.47	21.68	2.75	10.70	0.64	65.88	9.05	0.24
<i>Cladophora</i>	2.77	17.31	2.54	8.32	18.47	35.14	27.54	7.10
<i>Crustaceæ</i> :								
<i>Diaptomus</i>	10.38	64.87	8.01	...	8.58	12.60	5.94	...
<i>Cyclops</i>	9.57	59.81	19.80	...	10.07	4.58	5.74	...

PART II. ASH FREE

<i>Cyanophyceæ</i> :								
<i>Microcystis</i>	9.30	58.13	4.92	...	2.28	34.67		
<i>Microcystis</i>	9.68	60.55	2.79	5.19	0.27	36.39		
<i>Microcystis</i>	6.65	41.60	2.90	...	0.68	54.82		
Chiefly <i>Microcystis</i>	9.05	56.61	5.44	8.46		
<i>Anabæna</i>	8.91	55.68	1.20	5.18	0.68	42.44		
<i>Anabæna</i> and <i>Cœlo-</i> <i>sphaerium</i>	8.75	54.74	2.15	6.45	1.22	41.89		
<i>Aphanisomenon</i>	10.05	62.83	4.02	2.20	0.57	32.58		
<i>Aphanisomenon</i> and <i>Anabæna</i>	10.63	66.45	4.64	3.66	1.29	27.62		
<i>Lyngbya</i>	9.73	60.81	2.50	5.56	3.63	33.06		
<i>Lyngbya</i>	8.66	54.12	1.45	3.97	7.79	36.64		
<i>Diatomaceæ</i>	6.05	37.81	22.48	4.74	2.36	37.35		
<i>Chlorophyceæ</i> :								
<i>Volvox</i>	8.12	50.75	5.91	1.06	6.74	36.60		
<i>Spirogyra</i>	3.81	23.82	3.02	11.76	0.70	72.46		
<i>Cladophora</i>	3.77	23.56	3.45	11.32	25.14	47.65		
<i>Crustaceæ</i> :								
<i>Diaptomus</i>	11.03	68.93	8.51	...	9.12	13.44		
<i>Cyclops</i>	10.15	63.43	21.00	...	10.68	4.69		

TABLE 31

INORGANIC CONSTITUENTS OF THE ASH OF VARIOUS ORGANISMS
 Stated in Percentages of the Dry Weight of the Sample
After Birge and Juday

Organism	Ash	SiO ₂	Fe ₂ O ₃ and Al ₂ O ₃	P ₂ O ₅	CaO	MgO
<i>Cyanophyceæ:</i>						
Microcystis.....	4.31	0.13	0.84	1.18	0.92	0.63
Anabæna.....	7.17	0.95	1.27	1.21	1.42	0.70
Anabæna and Cœlosphærium.....	4.66	0.27	0.82	0.63
<i>Chlorophyceæ:</i>						
Volvox.....	6.28	0.24	0.80	2.50	1.10	0.93
Cladophora.....	26.54	7.10	1.80	0.32	3.26	1.62
<i>Crustaceæ:</i>						
Cyclops.....	5.74	1.43	2.32	0.78	0.75

TABLE 32

RESULTS OF ANALYSES OF FRESH WATER ORGANISMS
 Stated in Percentages of the Dry Weight of the Sample
As given by Various Investigators

Authority	Organism	Nitrogen	Crude Protein (N × 6.25)	Ether Extract	Crude Fiber	Ash	Silica
Whipple and Jackson	Microcystis (Clathrocystis)	8.30	51.87
	Anabæna	9.60	60.00
	Asterionella	2.20	13.75	57.52	49.48
	Spirogyra	4.50	28.12
Hyams and Richards	Oscillatoria (young)	9.00	56.25	4.50	1.46
	Oscillatoria (fully grown)	7.90	49.38	6.40	2.90
	Oscillatoria	9.19	51.19	2.43	...	7.09	...
Turner Brandt	Peridiniæ (chiefly Ceratium)	2.03	12.68	1.30	41.50	5.20	...
	Diatoms (chiefly Chætoceros)	1.56	9.75	2.80	...	65.20	54.50
	Freshwater Copepoda	9.16	57.25	6.01	5.54	9.21	...
Volk	Eurytemora	...	78.47	6.20	11.07	4.24	...
	Bosmina	...	77.82	10.66	8.21	3.30	...

Ash is produced in greatest amount from the diatoms, this being chiefly silica. Cladophora, a chlorophyllaceous organism, contains nearly as much ash as the diatoms; much of it is silica. The results in Table 31 show a considerable variety of inorganic constituents in the

ash, including silica, iron, alumina, phosphorus, calcium and magnesium. Of the forms given *Cladophora* is the highest in calcium, magnesium and iron content, seemingly possessing unusual ability to abstract these elements from the water.

Further information of this character is needed before many general conclusions can be drawn regarding the peculiarities and needs of particular genera or groups of organisms.

Environment. — Although food is a prime necessity for plankton life, it must be remembered that there are other factors that exercise modifying influences upon growth and reproduction. The previous chapter stressed the importance of the physical environment. Chemical conditions other than the mere presence of proper pabulum must likewise be taken into account. For instance, the hydrogen ion concentration of different waters, and of the same water, varies for one reason or another. The present state of our knowledge does not make possible a proper estimate of the effect of different hydrogen ion values upon the growth of microscopic organisms. It is well known that hydrogen ion concentration will change with fluctuating plankton growth, but it is not always clear to what extent the change bears a causal relationship to the growth or appears merely as an effect of growth. It is reasonable to expect, however, that the absorption of food and its ultimate utilization may be considerably affected by hydrogen ion concentration. Such is the case with the bacteria.

Another condition, not yet properly appraised, is the concentration of substances other than those needed for food. Do these sometimes stimulate cell activity? At what concentrations may they inhibit vital processes? There are many such conditions connected with the chemistry of lakes, the significance of which is obscure but compelling of recognition.

A Lake as a Closed Community. — Dr. Birge has well said:

The inhabitants of an inland lake form a closed community in a stricter sense, perhaps, than the term can be applied to any other non-parasitic assemblage. The number of species living under these conditions is small and closely similar in different lakes. Only small additions are made to the food supply from without and these come slowly. The lake is dependent on its own stock of green plants for the stock of organic matter available for food of other organisms; and the possible amount of green plants is limited by the raw material supplied for photosynthesis from the lake itself. The critical factor then, in the economy of a lake with small in- and out-flow of water, is the provision for the vertical circulation of the water in the lake. But this circulation is very imperfectly effected at best, and is often wholly absent for most of the water.

All of these factors co-operate to produce an annual cycle in the distribution of the dissolved gases, whose fundamental features are the same, but whose details differ endlessly in different lakes.

DISSOLVED OXYGEN AND FREE CARBON DIOXIDE

Natural waters may contain in solution all the gases of the atmosphere and in addition others resulting from bacterial activity, such as methane, carbon monoxide, and hydrogen sulphide. Of these, dissolved oxygen and carbon dioxide exert an important influence upon the growth of microscopic organisms, oxygen being necessary for respiration and carbon dioxide for nutrition of plant cells. There are many intimate relations between these gases and the organisms that produce and consume them.

Sources of Oxygen and Carbon Dioxide. — The principal sources of dissolved oxygen in the water of lakes are the atmosphere and the process of photosynthesis that takes place in green plants. Oxygen production is common to the minutest of these, the algæ. The establishment of this phenomenon quite fittingly belongs to Priestley, the English chemist, who was the first to announce the discovery of oxygen. Clean water in contact with the air is normally fully (100 per cent) saturated with dissolved oxygen. Water containing growing plants will, in the absence of pronounced bacterial activity, be fully saturated; oftentimes it may be supersaturated. Under conditions of heavy supersaturation much of the oxygen is probably held within the plant cells and is liberated during chemical treatment of a sample, when the cells are disrupted.

Carbon dioxide comes from the decomposition of organic matter and the respiration of animals and plants. The lower strata of water and the bottom sediment are the most productive sources, for it is there that the dead bodies of plant and animal organisms and lifeless organic matter of all kinds settle and are decomposed by the bacteria. Only to a slight extent is carbon dioxide absorbed from the atmosphere. Sometimes, however, this source is important, particularly when thermal stratification prevents carbon dioxide reaching the upper water from the lower layers. Ground water usually contains more carbon dioxide than surface water, and when discharged into a lake it adds to the content of this gas.

Solubility of Gases in Water. — Gases are conveyed from the atmosphere to the interior of standing bodies of water by, (a) solution from the atmosphere, (b) convection currents, (c) diffusion through the body of water. The *rate* at which these forces operate is influenced by many factors, the principal ones being shown in the summary below. There are many complex relationships and these vary with the nature of the gas and its effect upon the density, surface tension and viscosity of the water.

SOLUTION OF ATMOSPHERIC GASES IN WATER

Gases are conveyed from the atmosphere by:

I. Solution, the rate of which is influenced by

1. Solubility of the gas, which is controlled by
 - a. The partial pressure of the gas in the atmosphere
 - b. The solubility constant of the gas
 - c. The temperature of the water
 - d. The salt concentration
2. Degree of under-saturation of the water
3. Humidity of the atmosphere
4. Agitation of the water surface, caused by
 - a. Wind action
 - b. Irregular velocity of flow
 - c. Other mechanical disturbances

II. Convection currents, the rate of which is influenced by

1. Temperature changes, which are caused by
 - a. Changes in air temperature
 - b. Evaporation of water
2. Changes in surface salt concentration, induced by evaporation
3. Agitation of the body of water
(As a result of influences under I, 4.)

III. Diffusion, the rate of which is influenced by

1. Concentration of the gas in the water
2. Temperature, which affects the viscosity

Rate of Solution. — In accordance with Dalton's law for gaseous mixtures the gases of the atmosphere each exert a pressure approximately proportional to the volume percentage of gas times the total atmospheric pressure. This pressure is known as the "partial pressure" of the gas; it is not affected by the presence of the other gases. The total solubility of atmospheric gases in contact with quiescent water follows Henry's fundamental law. This states that the concentration of a gas in a solution at a definite temperature will vary proportionally with the partial pressure of the gas above the solution. Inasmuch as the partial pressure is a function of the total pressure if the latter is doubled twice as much gas will be dissolved. The solubility of the gas must also be taken into account, so that Henry's law may be stated as $C = Kp$, where C is the concentration of the gas in the solution, p the partial pressure of the gas and K is a constant of solubility.

The solubility of oxygen in water at a temperature of 0° C., when the

water is exposed to an atmosphere of the dry gas under a pressure of 760 mm., is 49.29 cc. per liter, of carbon dioxide 1713 cc. per liter, of nitrogen 23.00 cc. per liter. The atmosphere consists of 20.94 per cent by volume of oxygen, .03 per cent of carbon dioxide and 78.09 per cent of nitrogen, but in water saturated with air at 0° C. oxygen constitutes 34.91 per cent by volume of the total dissolved air and nitrogen only 65.09 per cent. Carbon dioxide disappears from the gaseous phase owing to its union with water to form carbonic acid. The saturation value for water at 0° C. in contact with air is about 1.4 parts CO₂ per million, or 0.7 cc. per liter. These volumes vary relatively from those which might be expected with the respective partial pressures by reason of the different *solubility constants* of the respective gases. If the air or gas in contact with the water is not dry the volume of the gas dissolved will be reduced in proportion to the partial pressure of the water vapor present in the supernatant atmosphere, i.e., the vapor tension of water.

The *total solubility* of gases also varies in general with the temperature of the water, being greater in cold than in warm water. At 20° C. and with water exposed to a pressure of the dry gas equal to 760 mm. the total solubility of oxygen is 31.44 cc. per liter, of carbon dioxide 878 cc. and of nitrogen 15.54 cc. Compare these values with those given above for 0° C. When water is warmed there is a tendency for gases to be driven from it due to lessened solubility. It is well to point out that although a rise in temperature decreases *total solubility* it increases the *rate of solution coefficient*, i. e., the rate of solution per unit area exposed when the difference between saturation and the actual concentration is 1 cc. per liter of water.

Increasing *concentration of dissolved salts* decreases the total solubility from that in distilled water. For this reason brackish water and sea water contain, when saturated, smaller volumes of atmospheric gases than does fresh water under the same conditions.

The degree of under-saturation at any one time speeds up the rate of solution, which becomes proportional to the difference between saturation concentration and the actual concentration. Rate of solution is also affected by the *humidity*, being greater when the air is dry than when it contains moisture. Dry air accelerates evaporation, which cools the surface of the water and sets up convection currents that distribute the gas through the water. *Agitation* of the water surface increases the rate of solution by bringing unsaturated films of water into contact with the air.

Convection Currents. — Down to 4° C. any drop in air temperature is transmitted to water in contact with it, increases the density of the

water at the surface and causes it to sink, thereby setting up *convection currents* and mixing the upper and lower water. The effect is marked at night. *Evaporation* lowers surface temperatures, increases water density, and so induces convection currents, which may extend their influence for several feet. It also increases the salt concentration at the surface, and hence the density. Convection currents are then set up in an attempt to restore equilibrium. Agitation of the water destroys the temperature equilibrium and gives rise to convection currents. Whatever may be the cause of the latter they promote a thinning of the surface film and allow readier passage to the gas.

Diffusion. — Diffusion is an important agency in the solution of gases and varies greatly with different gases. The *rate of diffusion* in water for a gas is mainly influenced by the *relative concentration of the gas* in the different water strata and by the temperature. It is directly proportional to the *difference in concentration*. Temperature is a factor by reason of the lowered viscosity of the surface film that results from increase in temperature; the film becomes thinner, permitting the gas to pass through it faster. Oxygen is only moderately soluble in water and diffusion is not a major factor in its transfer beyond the surface layers. The slow rate of its distribution in a lake by this means is illustrated by the computations of Hufner. He estimated that over a million years would be required to saturate by diffusion with oxygen from the air the waters of the Bodensee, which is 250 meters deep, were this lake deprived of its existing store of the gas.

Solubility of Oxygen. — The solubility of oxygen in water at different temperatures when the latter is exposed to an atmosphere containing 20.9 per cent of oxygen at a pressure of 760 mm. including pressure of water vapor, is given in Table 33. For elevations above sea-level 1 per cent should be deducted for every 270 feet of elevation. In comparing results expressed in parts per million by weight, i.e. milligrams per liter, with those expressed in cubic centimeters per liter it is convenient to remember that 1 cc. of oxygen, at 760 mm. pressure and 0° C., weighs 1.4292 mg. For oxygen values in salt water the reader is referred to "Standard Methods of Water and Sewage Analysis." The amount of dissolved oxygen present at any one time may be expressed in three ways: (1) in parts per million (milligrams per liter), (2) in cc. per liter, (3) as the percentage of saturation. The third method requires the measurement of the temperature of the water; the result is expressed as the percentage which the *amount of gas present* is of the *maximum amount capable of solution* in distilled water at the same temperature and pressure; the latter can be obtained from Table 33.

TABLE 33

AMOUNT OF DISSOLVED OXYGEN IN WATER AT DIFFERENT TEMPERATURES
Exposed to an atmosphere containing 20.9 per cent of oxygen under a pressure
of 760 mm. including pressure of water vapor.

Temp., °C.	Parts per Million	Cubic Centimeters per liter (at 0° C. and 760 mm.)	Temp., °C.	Parts per Million	Cubic Centimeters per liter (at 0° C. and 760 mm.)
0	14.62	10.23	16	9.95	6.96
1	14.23	9.96	17	9.74	6.82
2	13.84	9.68	18	9.54	6.68
3	13.48	9.43	19	9.35	6.54
4	13.13	9.19	20	9.17	6.42
5	12.80	8.96	21	8.99	6.29
6	12.48	8.73	22	8.83	6.18
7	12.17	8.52	23	8.68	6.07
8	11.87	8.31	24	8.53	5.97
9	11.59	8.11	25	8.38	5.86
10	11.33	7.93	26	8.22	5.75
11	11.08	7.75	27	8.07	5.65
12	10.83	7.58	28	7.92	5.54
13	10.60	7.42	29	7.77	5.44
14	10.37	7.26	30	7.63	5.34
15	10.15	7.10			

Solubility of Carbon Dioxide. — Carbon dioxide dissolves readily in water, forming carbonic acid which is largely dissociated in dilute solutions. As shown in Table 34 the amount that remains in solution at a given temperature is dependent upon the partial pressure of CO_2 in the atmosphere over the solution. In the open air the partial pressure is low, and water exposed to the open air in drops normally contains 1 to 2 parts per million of dissolved carbon dioxide. This dissolved carbon dioxide is sometimes called free carbon dioxide, to distinguish it from that present in bicarbonates and carbonates. The air in dug wells often contains considerable carbon dioxide; consequently their waters frequently contain large amounts of this gas as a result of the increased partial pressure. Other ground waters, too, often contain appreciable quantities of carbon dioxide. This originates in the ground air through which the water passes. The upper layer of soil known as the "zone of living earth" is often rich in decomposing organic matter which discharges carbon dioxide into the pores of the soil, whence it is absorbed by percolating water.

TABLE 34

SOLUBILITY OF CARBON DIOXIDE IN WATER

(Compiled from Sutton's Volumetric Analysis and Fox's paper in the Transactions of the Faraday Society, September, 1909)

Temperature, Centigrade	CC. per Liter	Parts per Million for Stated Partial Pressures of CO ₂ in the Atmosphere				
	1 part per 10,000	1 part per 10,000	4 parts per 10,000	6 parts per 10,000	8 parts per 10,000	
0	.1713	.34	1.4	2.0	2.8	
4	.1473	.29	1.2	1.7	2.4	
8	.1283	.26	1.0	1.5	2.1	
12	.1117	.22	.9	1.3	1.8	
16	.0987	.19	.8	1.2	1.6	
20	.0877	.17	.7	1.0	1.4	
24	.0780	.15	.6	.9	1.2	
28	.0780	.15	.6	.9	1.2	

Collection of Samples for Gases. — Dissolved oxygen and carbon dioxide in natural waters are seldom found in amounts that represent conditions of equilibrium with the atmosphere. It is, therefore, necessary to employ careful methods in sampling; air must not bubble through the sample during collection, for it may either contribute gases to, or expel them from, the water. Analysis should be made at the time of collection.

Dissolved Oxygen Apparatus. — Samples for oxygen determination are best collected in clear glass bottles fitted with ground stoppers and holding approximately 250 cc. Water from taps, or other sources under pressure, should be drawn through glass or rubber tubing attached to the tap and extending to the bottom of the bottle. Adequate flushing should first remove any air that is entrapped in the pipes. The water is then allowed to overflow until there is a displacement equal at least to the volume of the bottle. Reagents are added immediately following collection and the determinations made in accordance with standard procedure.

In the field, where samples must be taken from lakes, reservoirs and streams, some special form of apparatus must be used for collection. The simplest type, which has long been used, consists of two bottles arranged as shown in Chapter IV, Fig. 7, for Hale's sampling apparatus. In this a small bottle is clamped to a larger weighted bottle having two

to four times the volume. Connection between the two is established by glass and rubber tubing passing through 2-hole rubber stoppers. Water enters the small bottle through an open glass tube that runs to the bottom, while air leaves the large bottle through a tube of similar or slightly larger size, the flow being induced by reason of lower water pressure upon this than upon the tube leading to the small bottle. When the latter is full, water from it siphons over to the bottom of the large bottle until this, too, is full and all air is expelled from the apparatus. The last portion of water to enter the apparatus is that in the small bottle. It is a true sample of the water at a fixed depth.

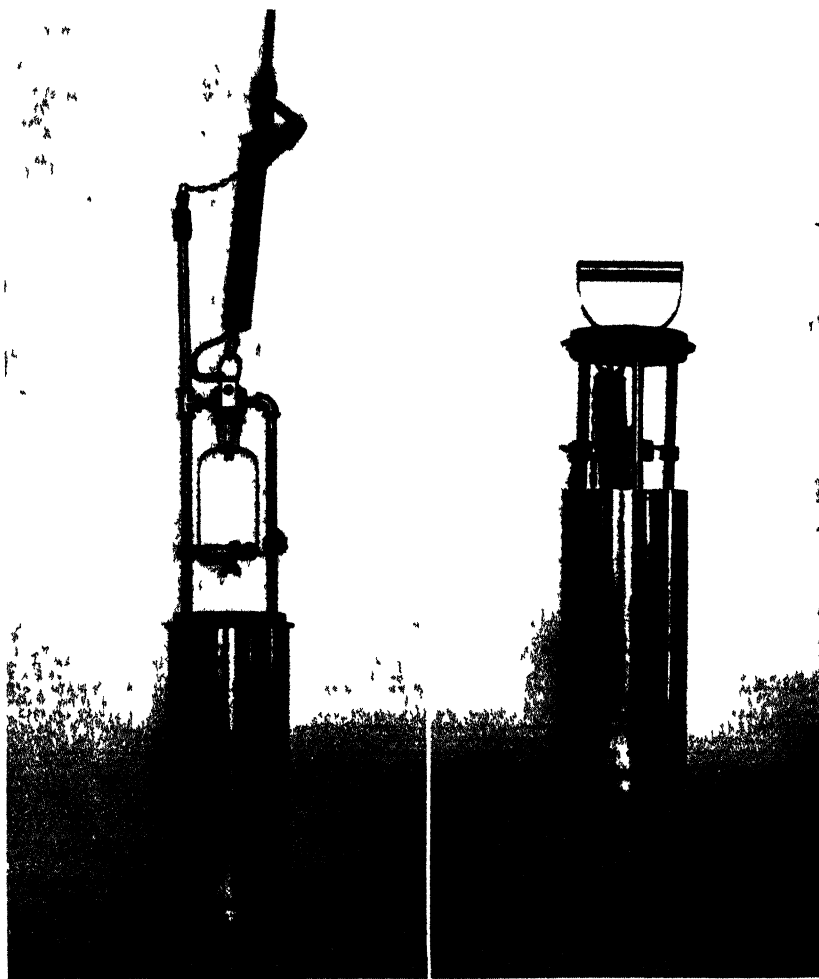
There is no stopper to be pulled with this apparatus; it starts filling immediately upon being submerged. The portion of water in the large bottle is, therefore, more or less a composite sample from the surface down.

Rugged Types of Apparatus. — Another apparatus, more compact and and rugged, which does not fill until a stopper is released at the desired depth, is that shown in Fig. 55. It is in use at the Laboratory of Sanitary Engineering, Harvard University. All parts of the frame and the lower container are of brass. The bottle has a capacity of about 250 cc. and rests upon a sliding platform held by set screws. It is sealed by pushing up against a rubber ring that is perforated to allow passage of the inlet tube. Concentric with the outside of this tube is an outlet leading through the solid brass head to the side tube on the right which communicates with the lower container. The outlet from the latter is through the tube at the left. This is carried about 8 inches above the inlet port shown in the brass head and is sealed by a small rubber stopper that is unseated at the desired depth by a pull on the rope. This operation puts tension on the coiled spring and jerks out the stopper. Air escapes from the tube and water enters the small bottle, first filling it and then the larger container. The cover of the container is threaded; to it is attached the upper framework. The whole may be unscrewed, reversed and fitted into the container to facilitate transportation. The figure shows this adaptation.

Any sampling apparatus that is lowered with one port open, while the other is closed by a stopper that is released at a desired depth, is exposed to the error of being partly filled with water during the lowering process. Compression of the air in the apparatus by the weight of the overlying water column allows water to enter. The amount of this error will depend upon the depth of submersion, the size of the open port and the elapsed time between casting the apparatus overboard and releasing the stopper of the second port.

In an apparatus like that pictured in Fig. 55 the small sample bottle

will first tend to be filled during lowering; after release of the stopper the water that enters will be flushed out into the larger container. At all ordinary depths the sample bottle will hold a true sample of the



Ready for Use.

Being Assembled for Transportation.

FIG. 55. — Sampling Apparatus for Dissolved Gases.

water at the desired depth; the water in the container, however, will be more or less composite of the total depth of the water column.

The larger container can be made to yield a true sample from the fixed depth if both ports are kept closed until lowering is completed and they are then simultaneously opened. An apparatus designed to

do this has been described by Dr. F. Ruttner of the Lunz Biological Station. It consists of a glass sample bottle and metal container, the latter having two ports that are both sealed by a single disk. The disk is held in place by two clips, or dogs, that fall back and release the disk when a messenger is sent down the rope. Water does not enter the apparatus until the release of the disk frees both ports simultaneously.

Another rugged type of oxygen sampler is that used by the Illinois State Water Survey and described in Bulletin No. 16 of the Survey,

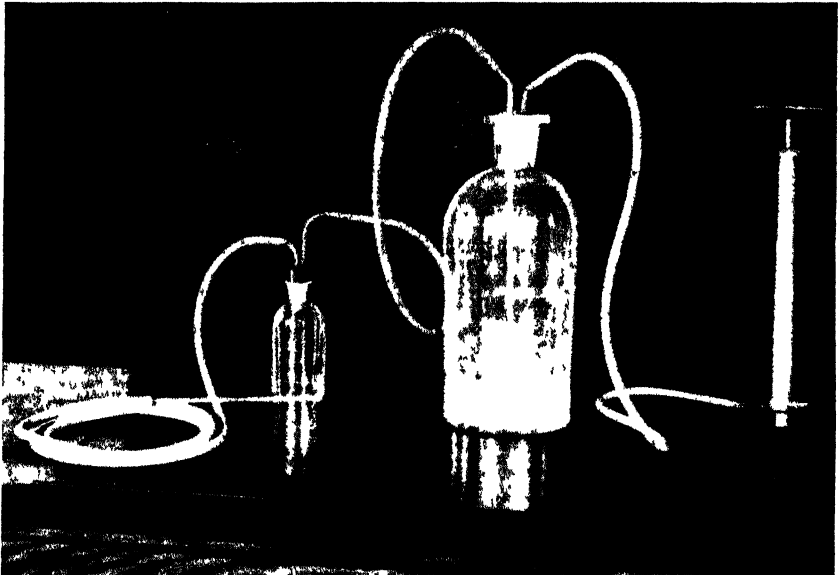


FIG. 56. — Sampling Device Employing Suction.

1918 to 1919, page 197. The apparatus consists of a weighted copper box $3\frac{1}{2}$ inch square by 10 inches deep. The cover is clamped to the box and carries on the inside a clamp for holding the sample bottle, which is suspended in the box. Inlet and outlet tubes of brass pass through the cover. No stopper is used. Filling starts as soon as the apparatus is submerged. The bottle first fills, then overflows until the whole box is filled.

Shallow Depth Apparatus. — Figure 56 shows an assembly of apparatus much used by the Laboratory of Sanitary Engineering, Harvard University. It enables samples to be taken from shallow water courses, from beneath the ice and from points beyond reach from the shore. The inlet tube is shown at the left and may be of any convenient length. The tube discharges at the bottom of the small bottle. A second tube

is carried to the bottom of the stopper of the small bottle and leads to the bottom of the large bottle. Another tube in the large bottle is flush with the bottom of the stopper and leads to a hand-operated suction pump, the latter being used to apply suction to the whole apparatus. The small bottle has a capacity of about 250 cc.; that of the large one may be 1, 2, or 4 liters. By the time the larger bottle fills a representative sample is collected in the smaller one for dissolved oxygen. The larger volume of sample can be used for microscopical and chemical examination.

If constructed with brass tubing instead of glass this apparatus becomes less fragile.

Carbon Dioxide Samples. — Owing to the ease with which carbon dioxide escapes from water, particularly when present in considerable quantities, it is necessary to collect a special sample for this determination and to make the analysis at the point of collection. In the case of surface waters where the carbon dioxide content is nearly in equilibrium with the air these refinements are sometimes omitted, but if aëration of the sample occurs there is usually danger of disturbing the gas content. Whenever there is a deficiency in the amount of the gas necessary to combine with normal carbonates aëration will produce high results. The usual error, however, contributes to loss of carbon dioxide and is in the direction of low results.

The determination should be made in accordance with "Standard Methods of Water Analysis." Actual titration is best carried out in a 100 cc. wide Nessler jar which is rotated with a centrifugal motion rather than being stirred with a glass rod.

Collection may be made after the manner described for dissolved oxygen samples. A portion of the water is then siphoned with the least possible disturbance into the titrating tube and the determination carried out.

Seasonal and Diurnal Changes in Dissolved Oxygen. — Lakes vary widely in their individual characteristics, in their exposure to climatic and meteorological changes and in the nature of the material carried in their waters. Consequently they manifest considerable differences in the amount and distribution of dissolved oxygen. In discussing some of the more general phenomena attending the variations of oxygen content in those deeper bodies of water that exhibit seasonal stratification and circulation it should be remembered that there are many modifications of, and departures from, the general statements made.

Let it be recalled that warm water will hold in solution less oxygen than cold water. Water saturated at summer temperature — say 20° C. — actually contains less oxygen, expressed in parts per million

or cubic centimeters per liter, than does water 65 per cent saturated at 0° C.

Normal Conditions. — In lakes and reservoirs used for public water supply the water above the transition zone is usually saturated with oxygen. This is principally due to the constant circulation of the water which continually brings it in contact with the air, and, also, to the growth of the phytoplankton and other hydrophytes. In the stagnation zone there is usually a depletion of the oxygen. The condition is most marked during the season of maximum temperature; it likewise prevails during winter stagnation, but in a diminished degree. If the amount of organic matter at the bottom is large, so that decomposition is active, the oxygen may be nearly or completely exhausted. Usually there is a gradual reduction within and below the transition zone. Sometimes, however, the change is very sharp. Thus in Irondequoit Bay, near Rochester, N. Y., on August 8, 1912, analyses showed the following percentages of saturation with dissolved oxygen at different depths:

TABLE 35
DISSOLVED OXYGEN IN IRONDEQUOIT BAY

Depth in Feet	Temperature °F.	Per Cent of Saturation
0	69.8	100.0
27	63.6	80.0
28	61.5	12.1
29	60.5	2.2
30	59.5	1.5
36	52.0	0.0
75	45.3	0.0

Figure 57, copied from the excellent report of Birge and Juday on "The Dissolved Gases of the Inland Lakes of Wisconsin," presents data that are typical for oxygen conditions in fairly deep lakes.

In the winter, when the surface is frozen, the oxygen supply from the air is cut off; at the same time the photosynthetic processes are at a low ebb, partly because of the cold and partly because the amount of radiant energy reaching the water is less. Yet respiration and decomposition continue, although these processes also are reduced in activity. The result is that beneath the ice the oxygen in the water tends to diminish. It is seldom greatly reduced unless the bottom of the lake is foul and decomposition excessive. In fact an excess of oxygen from

plankton activity is often noted in the upper waters beneath the ice if clear weather prevails and there is an absence of heavy snow to exclude light. Winter conditions are apt to vary greatly; the differences are due to the weather.

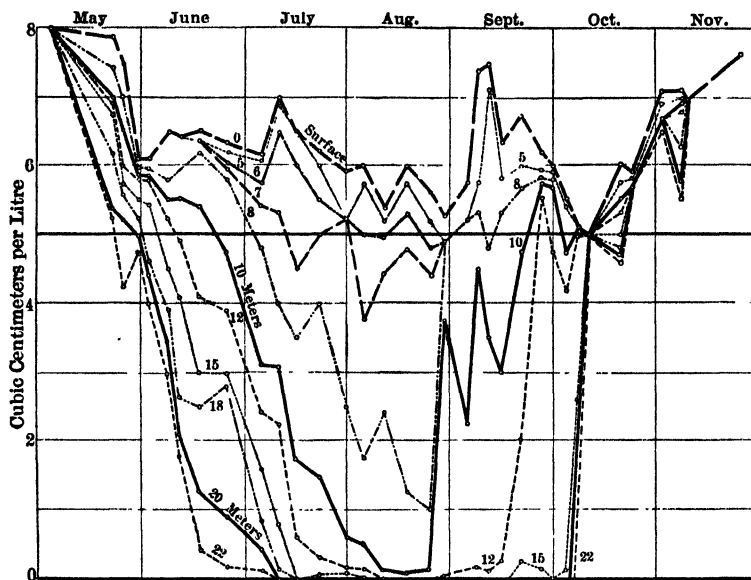


FIG. 57. — Dissolved Oxygen at Different Depths in Lake Mendota.
After Birge and Juday.

At the times of the spring and fall overturn the water is usually well aerated from top to bottom, although at the beginning of the overturn saturation values for oxygen frequently fall below 100 per cent, due to mixing of the lower, stagnant, oxygen-free water with the upper layers.

Decomposition in the Upper Zones. — Decomposition and oxygen depletion are most active in the zone of stagnation but they are not uncommon in the upper waters. The decay of algæ in a reservoir, following exhaustion of the food supply, may reduce the oxygen, even at the surface, faster than it is absorbed from the air. In shallow bodies of water the decay of algæ and organic matter may cause a depletion of oxygen sufficient to kill fish. An example of such an occurrence is given on page 209. Sometimes the depletion is noted in the upper part of the transition zone, the oxygen content being higher both above and below this depth and producing a notch on the plotted curve for oxygen. The curves of October 9 and 23, Fig. 59, illustrate in a moderate way this condition, which was probably brought about by the decay of dead

cells of algæ, the precipitation of which to the bottom had been arrested by the colder, denser water encountered at the transition zone.

Super-saturation. — The activities of those organisms which tend, by virtue of their chlorophyll content, to keep water saturated with oxygen are sometimes stimulated to a point where super-saturation results. Carbon dioxide is absorbed as food and resolved into its two elements, the carbon being retained by the cell and most of the oxygen liberated in the water. This most often occurs in the upper waters. The effect is not cumulative to a great extent, owing to the circulation that is maintained by wind action and convection currents, both of which promote contact of the water and the air with consequent loss of oxygen. The excess of oxygen will not greatly exceed that which is produced in a single day by the organisms. Birge found in Lake Mendota in 1908 that the maximum daily increase did not exceed 1.8 cc. per liter. This occurred at a depth of 1.5 meters. The maximum saturation was 175 per cent at this depth and at the surface 149 per cent.

Higher saturation figures than these are frequently obtained for the transition zone. Wind action seldom disturbs the water of this zone, convection currents are absent, diffusion at best is a slow process and decomposition of the plankton is slight during periods of active oxygen production. In clear waters chlorophyll-bearing organisms may find at this depth an abundant store of carbon dioxide and sufficient light to promote photosynthesis. The result is a release of oxygen which has been found in some cases to exceed 300 per cent of the saturation value. When brought to the surface such water may exhibit effervescence by reason of the release of hydrostatic pressure. There appears to be no close correlation between the depth at which the maximum concentration of oxygen occurs and that at which the greatest number of organisms is found.

Diurnal Changes. — The difference between day and night temperatures causes convection currents to be set up; if these are marked in their movement considerable differences are noted in oxygen values when algæ are numerous. At night oxygen production ceases and if the water in the zone of circulation is active there will be a distinct loss of the gas, perhaps amounting to from 1 to 3 cc. per liter. The morning oxygen curve will have a distinctly different profile from the evening curve. Decomposition and respiration continue at night and make further inroads on the oxygen content. When all these agencies are at work in small ponds that are heavily populated with green plants or with animal life, considerable oxygen deficiency may result over night, great enough to cause the death of fish. ‘

Seasonal Changes in Carbon Dioxide. — Under normal conditions of summer weather the vertical distribution of free carbon dioxide in lakes and reservoirs is the reverse of that of oxygen; it is low at the surface because the partial pressure of the gas in the atmosphere is low, and high at the bottom because of bacterial activity. Where photosynthesis is not taking place to any extent the water near the surface usually contains one to two parts per million of carbon dioxide. If decomposition is going forward in this zone the amount will be somewhat greater. If, however, green plants are present and food building is in progress the gas may be entirely absent and the water rendered

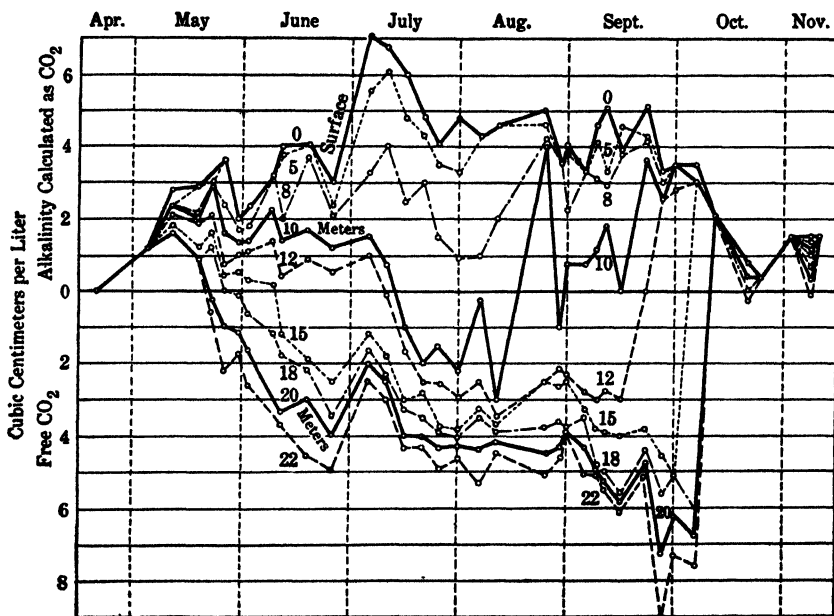


FIG. 58. — Carbon Dioxide in Lake Mendota at Different Depths.
After Birge and Juday.

alkaline to phenolphthalein by production of normal calcium carbonate. Lakes with large shoal areas, which allow bottom decomposition near the shore at depths above that of the transition zone, will, in general, contain more carbon dioxide in their upper waters than lakes with steep shore lines.

The transition zone will contain more carbon dioxide than the zone of circulation, for, here, there is usually a decrease in food building and an increase in rate of decomposition. The zone of stagnation gradually increases its content of the gas as summer advances. Temperatures favor bacterial activity, respiration of animal forms is common; photo-

synthesis is absent; mixing with the upper waters is prevented by stratification. The cumulative increase may amount to 25 and even 50 cc. per liter. The vertical distribution of carbon dioxide in a deep lake is shown in Fig. 58 taken from Birge and Juday.

Autumnal Changes. — At the approach of the fall overturn the great store of carbon dioxide that has been locked up in the lower layers is gradually released. Thermal changes promote deeper and deeper circulation. Finally the entire body of water is in circulation. Much free carbon dioxide is then brought to the surface and provides food for the phytoplankton. The autumnal increase in the plankton growth usually has its inception before the overturn is complete, being coincident with the first distinct depression of the transition zone.

Winter Conditions. — Autumnal circulation causes a loss of considerable carbon dioxide through aëration and through combination with normal carbonate of the surface layers. By the time ice forms there is a uniform concentration of 1 or 2 cc. per liter, sometimes less. The uncertain and unstable weather of winter affects the vertical distribution of the gas in different ways. Thaws and mild weather promote circulation and a uniform concentration of gas. Severely cold weather causes stratification of the water and accumulation of carbon dioxide at the bottom. This never approaches, however, the amount found in summer. Sunlight promotes photosynthesis and brings about a depletion of the gas in the water near the ice.

Example of Observations on Dissolved Gases. — A series of studies made upon Fresh Pond, Cambridge, Mass., in 1913 to 1914 illustrates typical changes in temperature, dissolved oxygen and free carbon dioxide that may be expected in the course of an annual cycle. Graphical representation of this work will be found in Fig. 59. Fresh Pond has a capacity of about 1400 million gallons, a small catchment area, and was fed at the time by soft, colored water from impounded sources. It has a maximum depth of 55 feet and considerable areas with a depth of 10 to 15 feet. (See Fig. 102, Chapter XIII.) The bottom is foul with mud and organic ooze. About 12 million gallons per day were being withdrawn from the Pond for water supply purposes.

The first observations, diagram of Oct. 9, show typical fall conditions with temperature stratification, carbon dioxide low in the zone of circulation and increasing rapidly to 18 parts per million at the bottom, oxygen to the amount of saturation in the upper 10 feet, then a sharp decrease, and entire absence below 30 feet. The notch in the oxygen curve below 20 feet illustrates reduction due to decomposition of dead algal cells that were arrested in their fall to the bottom by denser water below this point. *

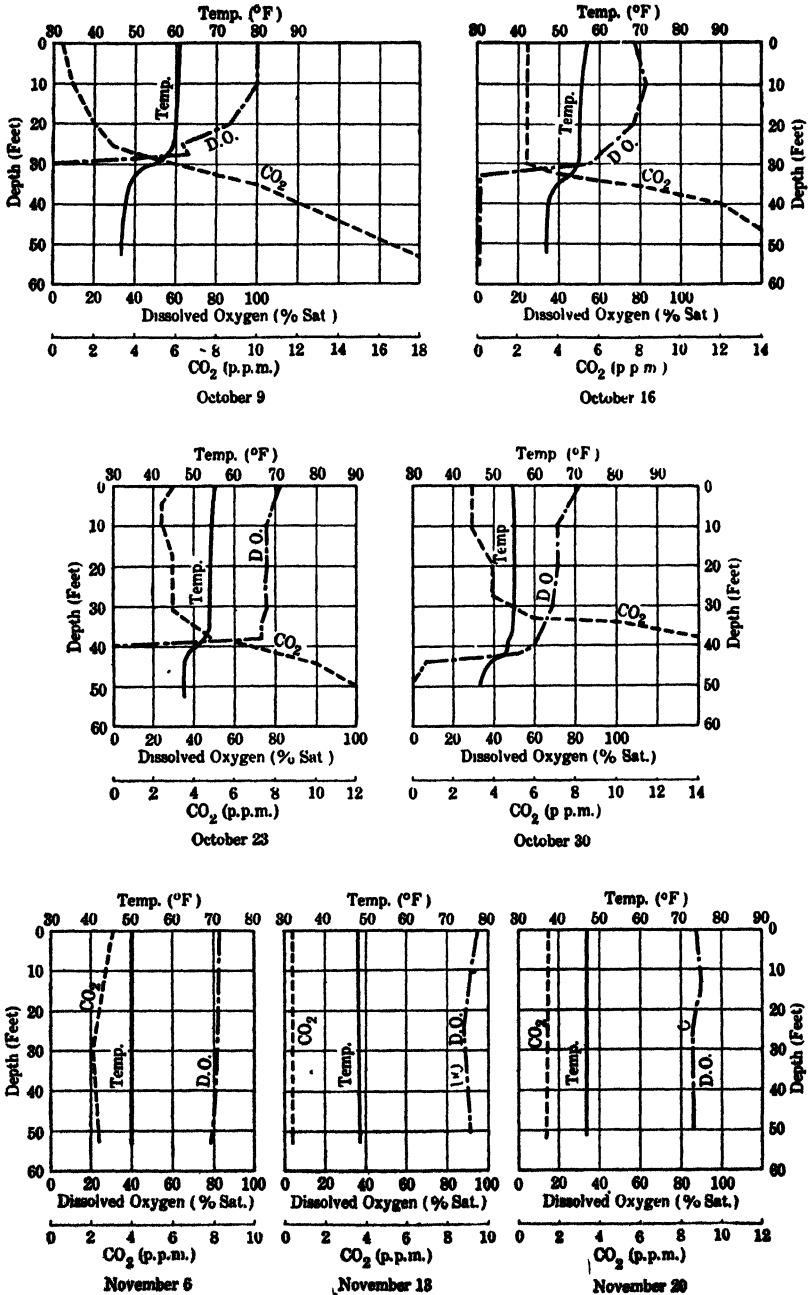


FIG. 59. — Studies of Temperature and Dissolved Gases, Fresh Pond, Cambridge, Mass. October, November 1913.

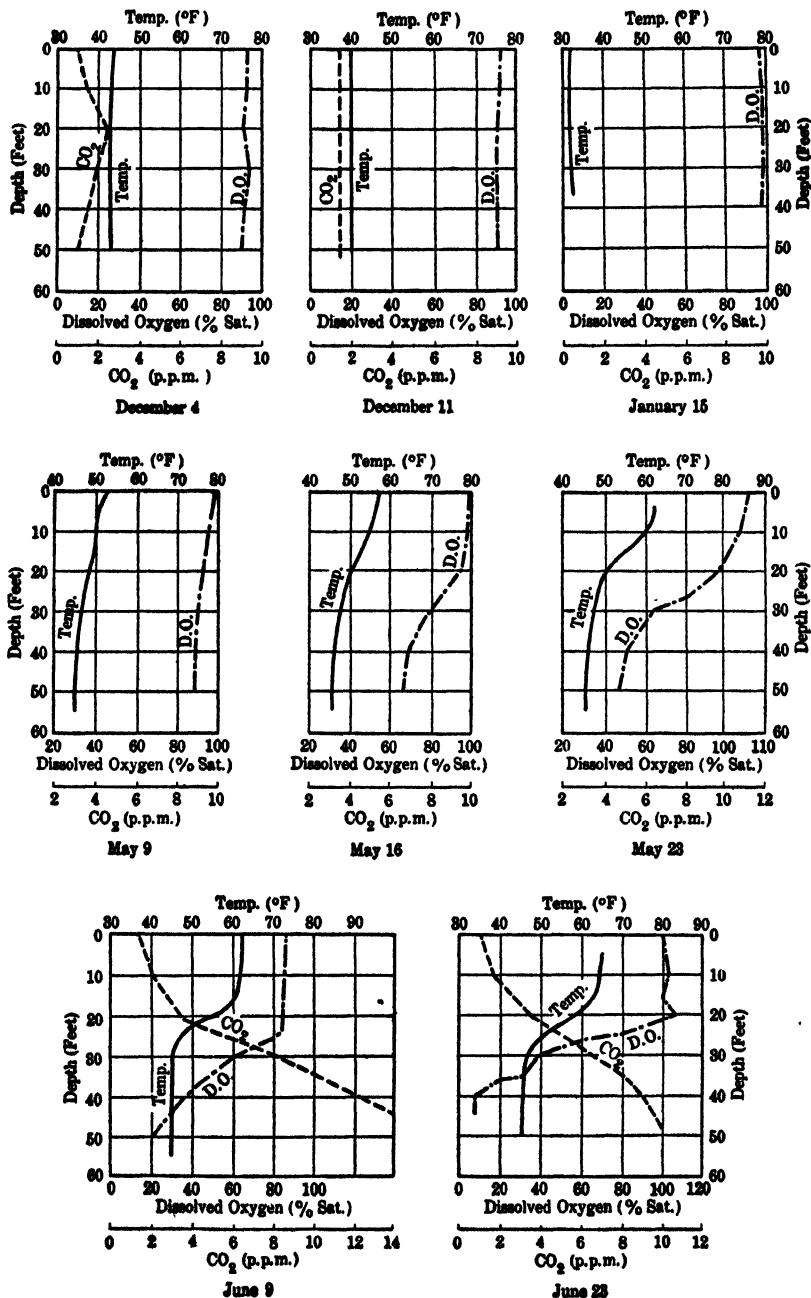


FIG. 59. — Studies of Temperature and Dissolved Gases, Fresh Pond, Cambridge, Mass. December 1913 to June 1914.

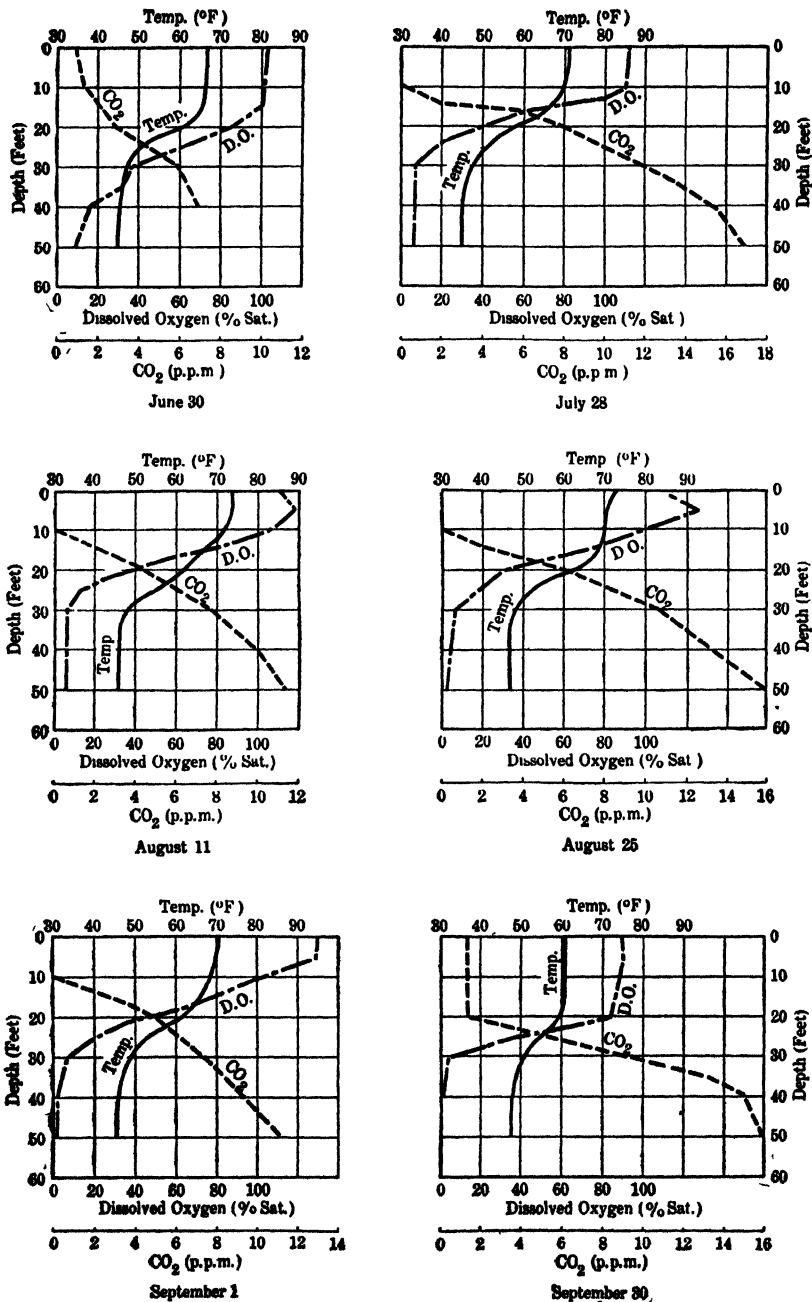


FIG. 59. — Studies of Temperature and Dissolved Gases, Fresh Pond, Cambridge, Mass. June to September 1914.

Proceeding chronologically, it is seen that the transition zone was gradually depressed by colder weather. Gas conditions were disturbed by increasing circulation, oxygen falling below the saturation value in the upper zone and carbon dioxide increasing in amount. By Nov. 6 the pond was in active vertical circulation following the fall overturn and on Dec. 11 the plotted values for temperature and gases fell on a straight line. On Jan. 15 there was a slight indication of winter stagnation.

There were no observations from Jan. 15 to May 9 when the first indications of summer stratification were apparent. As the season advanced the diagrams show more closely defined limits for the transition zone and continued depletion of oxygen in the lower waters. The carbon dioxide increased rather irregularly and oxygen values varied. On June 23 there was an excess of this gas at the 20 foot depth, probably due to the activity of microscopic organisms. By July 28 all the carbon dioxide in the upper 10 feet was used up by growing plants. The release of oxygen produced supersaturation. These conditions held good until September 30 when autumnal circulation had again started, as is shown by depression of the transition zone with respect to its position of September 1. Maximum oxygen concentration at this time occurred in the upper 15 feet of water.

Relation of Dissolved Gases to Algæ. — The best discussion of this subject is to be found in a paper by Dr. Charles O. Chambers published in the twenty-third annual report of the Missouri Botanical Gardens, issued Dec. 18, 1912. Chambers not only compiled data from various foreign laboratories but carried on a series of experiments made in the lagoons of the botanic gardens at St. Louis, where blue-green algæ were growing. Of especial interest is his observation that on clear, sunny days the water became supersaturated with dissolved oxygen, while on cloudy days the percentage of oxygen fell below saturation, sometimes as low as 40 per cent. In general the carbonic acid increased as the oxygen decreased, but this reciprocal relation did not always hold. A similar fluctuation in gaseous content also occurs between day and night according to authorities quoted. Another interesting finding was that aëration tends to the formation of individual cells, while in poorly aërated water there is a tendency for organisms to form colonies and filaments.

Chambers has summarized the results of his findings as follows:

1. There is an intimate and mutual relation between the algæ and submerged aquatics in a body of water and the gases dissolved in that water. They fluctuate together.

2. Air, or its constituents, oxygen and CO_2 , are as essential to water plants as water is to land plants, and equally difficult to secure.

3. Warm and stagnant water is poorer in these essentials than colder water gently agitated by wind or currents.

4. Currents are especially beneficial to attached plants by renewing or removing these gases.

5. Some species demand more aëration than others. Some species are more tolerant of stagnant waters than others.

6. Filamentous forms with large cells and thin outer walls are best adapted to stagnant waters. Such forms predominate in warm, tropical fresh waters, which are poorly aërated.

7. The photosynthesis of rapidly-growing algæ and aquatic plants in a body of water may diminish or deplete the supply of CO_2 and increase the oxygen content beyond saturation.

8. In the absence of free CO_2 the plants may utilize the half-bound CO_2 of the dissolved bicarbonates, chiefly those of calcium and magnesium.

9. The process of photosynthesis may be so vigorous as to exhaust the half-bound CO_2 and render the water alkaline. By respiration and absorption of CO_2 from the air more bicarbonates may be formed. This serves as a mechanism for the conservation of CO_2 .

10. Waters rich in lime-carbonates are also rich in vegetation. Bog waters, containing humic acids, and, consequently, poor in carbonates of lime, are known to be poor in vegetation.

11. Stagnant water, on account of the large amount of CO_2 and the small amount of oxygen, favors the formation of colonies and filaments rather than of free individual cells.

12. Colonies and filamentous forms may be produced artificially with some plants, by increasing the amount of CO_2 or diminishing the amount of oxygen in the culture solutions.

13. Narrow, much-branched filaments are adapted to and produced by poorly aërated waters.

14. Aëration, or abundance of oxygen, apparently favors the formation of chlorophyll; and algæ are brighter green when well aërated.

15. The periodicity of spore formation is not readily influenced by aëration or gas content of the water. It seems to be more a matter of heredity.

Dissolved Gases and the Zoöplankton. — There is no well established relationship between the animal plankton and dissolved gases. Aside from the fact that oxygen is a requisite for the respiration of animal forms the gases found in water, both those produced by synthetic and by analytic processes, appear to exert little effect upon such animal life as can exist in lakes in the absence of photosynthetic reactions. Birge has observed that many protozoa are able to adapt themselves to an entire absence of dissolved oxygen and carry on their life processes as well under anaërobic as under aërobic conditions. He reports the same as true for some of the worms, *Anguillula*, *Limnodrilus* and *Tubifex*.

Higher forms of animal planktons, such as the rotifera and crustacea, have rarely been found in an entire absence of dissolved oxygen but are often numerous in concentrations of 0.1 to 0.3 cc. per liter, which amount is common to the layer of water just above the zone of total

depletion. These organisms are apparently sensitive to an entire lack of dissolved oxygen and migrate from such a zone. Experiments have shown that there is no sensitiveness toward the gases of decomposition such as carbon dioxide, methane, carbon monoxide and hydrogen sulphide, at least not toward such concentrations as are naturally produced. Neither does water supersaturated with oxygen exercise a detrimental influence upon these organisms.

Death of Fish at Newark, N. J. — In August, 1906, a large number of fish suddenly died in the lake at the Weequahic Reservation, Newark, N. J. This was investigated by Herbert B. Baldwin and the author; the results may be found in a report made to the Park Commission of Essex County, N. J., for that year.

TABLE 36
ANALYSES OF WEEQUAHIC LAKE WATER

Date	Depth, Feet	Temp., °F.	Dissolved Oxygen		CO ₂ , p.p.m.	Microscopic Organisms
			p.p.m.	Per Cent Saturation		S.U. per cc.
Aug. 21	0	82.0	52	16,350*
	1	31	
	3	3	
	6	3	
Aug. 22	0	79.0	4.9	59	
	1	1.8	22	
	2	1.0	12	
	3	0.7	9	
	4.5	0.6	7	
Aug. 24	0	75.5	5.2	61	
	3	4.2	50	
	5	75.0	4.2	49	
Aug. 27	0	77.5	15.0	180	0	
	3	15.0	180	0	
	5	77.0	13.2	157	0	

* 12,500 units were *Clathrocystis* and 2700 were *Anabæna*. Both were in a state of disintegration.

The lake covered about 80 acres and had an average depth of between 5 and 6 feet, although in a few spots the water was 12 feet deep. The

site of the reservoir was a swamp in which the depth of mud and peaty matter varied from 2 to 10 feet. This mud was not removed when the reservoir was constructed. Aquatic plants, water weeds and filamentous algæ flourished in the lake and at times great masses of peat and stumps floated to the surface. In the summer heavy growths of blue-green algæ occurred.

On the night of August 19 twelve two-horse loads of dead fish were picked up on the shore and it was estimated that more than fifteen tons died in two days. The dead fish included bass, roach, sunfish, horn pout, suckers, eels, and a few carp. They varied in size from sunfish 2 inches long to black bass weighing 5 pounds. The investigation showed that the probable cause of the death of the fish was an almost complete exhaustion of oxygen which resulted from the sudden decay of the algæ, principally *Clathrocystis* and *Anabæna*, which had been growing in the lake. The analyses made by the investigation offered additional testimony to the disastrous consequences that attend a disturbed biological balance. Some of the results are given in Table 36.

It is interesting to note the return to conditions of supersaturation on Aug. 27, which was marked by a rapid growth of diatomaceæ.

UTILIZATION OF INORGANIC SUBSTANCES

Necessary as are the dissolved gases to the vital processes of the microscopic organisms, there are other important substances in water that are drawn upon to furnish food and energy. Among these are certain inorganic compounds.

Rôle of the Bicarbonates. — Nearly all natural waters, in the absence of plant growths, owe their alkalinity to the bicarbonates of calcium and magnesium. Alkalinity may be determined by the use of the indicator methyl orange, or erythrosine, with a standard acid and is expressed in terms of normal calcium carbonate (CaCO_3). Natural waters also contain free carbon dioxide and so are acid to the indicator phenolphthalein.

Effect of Removing CO_2 . — If green plants are present and food building is in progress, all the free carbon dioxide may be used up. Further, some of the carbon dioxide, or even all of it, may be taken from the bicarbonates, leaving normal carbonates of calcium and magnesium which are not very soluble. If the amount of bicarbonates was originally large there may be a precipitation of normal calcium carbonate brought about in this way. When carbon dioxide has been removed from bicarbonates the water becomes alkaline to phenolphthalein and the free carbon dioxide assumes a negative value. Bicarbonates and

carbonates can be present in water at the same time, as can bicarbonates and free carbon dioxide; normal carbonates and free carbon dioxide will not be found together. For factors used in the conversion of alkalinity results obtained with methyl orange and phenolphthalein, and of carbon dioxide results, the reader is referred to "Standard Methods of Water Analysis."

Action of Plant Organisms. — It is not uncommon during the growing season to find the circulation zone of lakes devoid of free carbon dioxide and deficient in bicarbonates. In fact the same condition is occasionally present beneath ice when light intensity is high enough to promote photosynthesis in the plankton. Supersaturated values for oxygen usually occur at the same time. Growths in the transition zone will sometimes produce a like effect. At such times the water is alkaline to phenolphthalein, due to the presence of normal carbonates.

Marked changes are noted at the same time in pH value, i.e. in the hydrogen ion concentration. Removal of free carbon dioxide from water causes a reduction of hydrogen ion concentration and, therefore, an increase in the pH value. The change is further promoted by release of normal carbonates when carbon dioxide from bicarbonates is utilized by plants. As a consequence, waters that are nearly neutral on the hydrogen ion scale and have a pH value around 7 may at times of abundant plant growth exhibit a reaction of pH 8 or higher.

Just what algæ and water plants are able to take away carbon dioxide from bicarbonates is not known. Possibly all of them can. It is believed that such water weeds as *Potamogeton* (pond weed), *Carex* (sedge), and *Batrachium* (water buttercup) draw heavily on the bicarbonates; it is also known that blue-green algæ, such as *Anabæna* and *Clathrocystis*, and diatomaceæ, such as *Asterionella*, will do the same.

Studies on Jamaica Pond. — A typical example of the way in which bicarbonates are drawn upon as food when carbon dioxide becomes scarce was afforded by observations made on Jamaica Pond in Boston. The results are given in Table 37.

Jamaica Pond is included in the Metropolitan Park District. It has an area of about 7.5 acres and is served by a small catchment area. The bottom is soft and covered with organic deposits. About May 15, 1925, it took on a pink tint which increased in a week to a copper red color. The cause of this was a prodigious growth of *Oscillatoria prolifica* in almost pure culture.

It is interesting to note how the organisms migrated toward the bottom of the pond in search of food. All the carbon dioxide was used up down to 15 feet and most of that in the bicarbonates down to 10 feet. Oxygen saturation was practically coincident with the dis-

appearance of carbon dioxide. The pH values varied markedly with the depth. The oxygen values near the surface were lower than the number of organisms would suggest, but it is probable that some oxygen was being used up by decomposition; the surface organisms had begun to disintegrate and bacteria were increasing.

TABLE 37
OBSERVATIONS ON THE WATER OF JAMAICA POND, BOSTON
May 22, 1925

Depth, Ft.	Temp., °F.	Micro-organisms, Standard Units per cc.	Dissolved Oxygen, Per Cent Saturation	CO ₂ in p.p.m.	H.I.C pH	Alkalinity, p.p.m. CaCO ₃			Bacteria per cc.	
						Total	Normal Carb.	Bicarb onate	37° C.	20° C.
0	67.5	386,000	118	0	7.8	18.5	18.0	0.5	12	850*
5	64.3	290,000	117	0	7.8	18.5	17.0	1.5		
10	58.5	184,000	100	0	7.7	18.5	13.0	5.5		
15	55.0	110,000	90	0	7.5	18.5	5.0	13.5		
20	51.0	96,000	79	2.0	7.4	18.5	0	18.5		
26	47.5	62,000	67	10.5	7.1	18.5	0	18.5		

* On May 20 the bacteria growing at 20° were 73 per cc.

Relation of Alkalinity to Plankton. — Hard-water lakes in which the content of bicarbonates is high contain a store of carbon dioxide not found in soft water although the amount of free carbon dioxide present in the upper waters may or may not be greater than in soft water. Such lakes may indirectly furnish for plant growth a large amount of carbon dioxide. This is taken from the air when bicarbonates have been largely changed to normal carbonates, for water containing much normal carbonate will absorb carbon dioxide more rapidly than water containing little or none.

There are factors other than the hardness of water that greatly influence the ability of a lake to promote plankton growth; when these are comparable a lake containing available food in the form of high bicarbonates will usually be found to promote the development of chlorophyllaceous plants to a greater extent than one low in bicarbonates. Birge and Juday in their studies of Wisconsin lakes found none which contained so little phytoplankton as many of the soft water lakes.

The desmids (a family of green algae) as a group seem, according to several workers, to prefer the acid conditions of soft-water lakes. A statistical study of Massachusetts lakes and reservoirs, made in 1900,

showed that of 10 lakes low in hardness not one contained an average number of protozoa as high as 1000 per cc., while of 11 lakes high in hardness all contained an average number of protozoa above 100 per cc., and 4 had more than 1000 per cc. Diatoms also favored those bodies of water which were high in hardness. It is probable, however, that it is the greater amount of free carbon dioxide accompanying the waters of higher hardness that stimulates the growth of organisms, rather than the salts of calcium and magnesium. Tables 38 to 41 present the results of this statistical study.

Inorganic Forms of Nitrogen. — Since nitrogen is essential to all living matter we naturally expect that organisms will thrive best in waters rich in that element. The transformations wrought upon organic nitrogen by the operation of the nitrogen cycle produce in turn ammonia salts or free ammonia, principally carbonate, and nitrites and nitrates. Of these three the nitrates represent nitrogen in the most available form for the food of such organisms as absorb their nourishment through the cell membrane.

Relation of Nitrates to Plankton. — Organisms that directly assimilate their food are stimulated in growth in such bodies of water as contain an abundant store of nitrates. Further, it has been found that growth does not depend upon an unusually high nitrate content at any one time if the sources from which this is produced are abundant, namely free ammonia and nitrite. It is not at all uncommon to find large pelagic and shore growths thriving in the presence of a few hundredths of a part per million of nitrate nitrogen, when it is maintained at that amount by new increments produced from oxidation of free ammonia and nitrites.

The studies of Massachusetts lakes, previously referred to and summarized in Tables 38 to 41 indicate that the diatomaceæ, chlorophyceæ, cyanophyceæ and protozoa were all more numerous in those bodies of water that contained the largest amounts of free ammonia and nitrates. Care must be taken, however, in judging these chemical conditions, not to mistake cause for effect. Free ammonia, for example, indicates organic matter in a state of decay, and instead of representing the food of organisms it may represent the decay of available food or the decomposition of the organisms themselves.

The true and intimate relations of mineral nitrogen and plankton growth have not been worked out. We do know that given other necessary food and conditions of environment the phytoplankton is stimulated, as is all plant growth, by the addition of nitrates to the water. We also know that nitrates tend to increase in cold weather after the growing season, and that free ammonia and nitrites decrease

TABLE 38

OCCURRENCE OF CYANOPHYCEÆ IN MASSACHUSETTS LAKES AND RESERVOIRS

Chemical Analysis (parts per 1,000,000)		Number of Lakes and Reservoirs in which the Cyanophyceæ are			
		Often above 1000 per cc.	Occasion- ally above 1000 per cc.	Usually between 100 and 500 per cc.	Below 100 per cc.
Color	0 to 30	2	4	12	11
	30 to 60	2	3	4	3
	60 to 100	3	2	1	7
	above 100	0	1	1	1
Excess of Chlorides, (above the normal)	0	2	1	3	3
	0.1 to 0.3	1	3	5	10
	0.4 to 2.5	1	5	8	9
	above 2.5	3	1	2	0
Hardness	0 to 5	0	2	1	6
	5 to 10	2	2	4	10
	10 to 20	2	5	7	5
	above 20	3	1	6	1
Albuminoid Ammonia (dissolved)	0 to 0.100	0	0	1	4
	0.100 to 0.150	0	3	6	6
	0.150 to 0.200	2	5	8	7
	above 0.200	5	2	3	5
Free Ammonia	0 to 0.010	0	2	1	10
	0.010 to 0.030	0	2	9	8
	0.030 to 0.010	3	5	6	4
	above 0.100	4	1	2	0
Nitrates	0 to 0.050	1	2	1	12
	0.050 to 0.100	3	4	10	10
	0.100 to 0.200	1	3	5	0
	above 0.200	2	1	2	0

TABLE 39

OCCURRENCE OF CHLOROPHYCEÆ IN MASSACHUSETTS LAKES AND RESERVOIRS

Chemical Analysis (parts per 1,000,000)		Number of Lakes and Reservoirs in which the Chlorophyceæ are			
		Often above 1000 per cc.	Occasion- ally above 1000 per cc.	Usually between 100 and 500 per cc.	Below 100 per cc.
Color	0 to 30	2	5	14	8
	30 to 60	2	4	5	1
	60 to 100	1	2	8	2
	above 100	0	0	2	1
Excess of Chlorides, (above the normal)	0	1	3	4	1
	0.1 to 0.3	1	2	11	5
	0.4 to 2.5	0	4	13	6
	above 2.5	3	2	1	0
Hardness	0 to 5	0	2	3	4
	5 to 10	1	4	8	5
	10 to 20	1	3	13	2
	above 20	3	2	5	1
Albuminoid Ammonia (dissolved)	0 to 0.100	0	0	2	3
	0.100 to 0.150	0	4	7	4
	0.150 to 0.200	2	5	12	3
	above 0.200	3	2	8	2
Free Ammonia	0.000 to 0.010	0	2	7	4
	0.010 to 0.030	0	1	13	5
	0.030 to 0.010	2	5	8	3
	above 0.100	3	3	1	0
Nitrates	0 to 0.050	0	2	8	6
	0.050 to 0.100	2	6	13	6
	0.100 to 0.200	0	2	7	0
	above 0.200	3	1	1	0

TABLE 40

OCCURRENCE OF DIATOMACEÆ IN MASSACHUSETTS LAKES AND RESERVOIRS

Chemical Analysis (parts per 1,000,000)		Number of Lakes and Reservoirs in which the Diatomaceæ are			
		Often above 1000 per cc.	Occasion- ally above 1000 per cc.	Usually between 100 and 500 per cc.	Below 100 per cc.
Color	0 to 30	12	4	9	4
	30 to 60	6	2	4	0
	60 to 100	6	1	5	1
	above 100	0	1	1	1
Excess of Chlorides (above the normal)	0	4	2	1	2
	0.1 to 0.3	8	1	8	2
	0.4 to 2.5	8	3	10	2
	above 2.5	4	2	0	0
Hardness	0 to 5	2	1	3	3
	5 to 10	7	4	5	2
	10 to 20	8	0	10	1
	above 20	7	3	1	0
Albuminoid Ammonia (dissolved)	0 to 0.100	2	0	2	1
	0.100 to 0.150	6	1	5	3
	0.150 to 0.200	8	6	7	1
	above 0.200	8	1	5	1
Free Ammonia	0.000 to 0.010	3	2	5	3
	0.010 to 0.030	6	1	10	2
	0.030 to 0.100	8	5	4	1
	above 0.100	7	0	0	0
Nitrates	0 to 0.050	3	3	5	6
	0.050 to 0.100	11	3	13	0
	0.100 to 0.200	6	2	1	0
	above 0.200	4	1	0	0

TABLE 41

OCCURRENCE OF PROTOZOA IN MASSACHUSETTS LAKES AND RESERVOIRS

Chemical Analysis (parts per 1,000,000)		Number of Lakes and Reservoirs in which the Protozoa are			
		Often above 1000 per cc.	Occasion- ally above 1000 per cc.	Usually between 100 and 500 per cc.	Below 100 per cc.
Color	0 to 30	5	2	20	2
	30 to 60	1	3	6	2
	60 to 100	2	2	8	1
	above 100	0	0	1	2
Excess of Chlorides (above the normal)	0	1	2	5	1
	0.1 to 0.3	1	2	13	3
	0.4 to 2.5	2	3	15	3
	above 2.5	3	0	3	0
Hardness	0 to 5	0	0	7	3
	5 to 10	3	0	12	2
	10 to 20	1	6	10	2
	above 20	4	1	6	0
Albuminoid Ammonia (dissolved)	0 to 0.100	0	0	4	1
	0.100 to 0.150	0	0	13	2
	0.150 to 0.200	5	2	12	3
	above 0.200	3	4	7	1
Free Ammonia	0 to 0.010	1	1	9	2
	0.010 to 0.030	1	1	13	4
	0.030 to 0.100	2	5	10	1
	above 0.100	4	0	3	0
Nitrates	0 to 0.050	0	1	12	3
	0.050 to 0.100	3	4	17	3
	0.100 to 0.200	3	2	3	1
	above 0.200	2	0	3	0

due to inhibited bacterial activity. In the spring ammonia and nitrites usually increase in advance of growing plant life. With the advent of temperatures favorable to their oxidation and to plant growth they may decrease, as will the nitrates which are directly available for food.

Silica. — The diatomaceæ are characterized by their utilization of silica to form siliceous cell walls. At times of their abundant growth silica is removed from the water in appreciable amounts and this is followed by an increase in the silica content of the lower waters, due to the precipitation of the dead cells and a solvent action of the water or its constituents upon the silica. The maximum silica content in the lower stagnant waters occurs just prior to the fall overturn.

Other Mineral Substances. — Little importance attaches to many mineral substances in connection with the development of the microscopic organisms, for the reason that little or no change is registered in the amount of these substances at times of maximum growth. On the other hand we know but little about some of the minor problems of nutrition and growth which may in the future be shown to depend upon factors now little appreciated, and which may lead to a better understanding of the laws governing the occurrence and distribution of microscopic forms of aquatic life. Iron, calcium, magnesium, the sulphates and the chlorides appear to be without stimulating or inhibiting effect in the amounts in which they are found in natural waters. Phosphorus is apparently utilized by the algæ; it has been observed to decrease in lake waters during seasons when microscopic plants were abundant in numbers.

Relation of Plankton to Excess of Chlorides. — “Excess of chlorides” is the difference between the amount of chlorides found in a sample of water and that found in uncontaminated water of the same region. Generally, it is a measure of the sewage contamination that the water has received. It is important to know whether this element of the analysis bears any relation to the organisms and whether one may rightly infer that a large growth of organisms in a reservoir is an indication of contamination of a water supply. In the Massachusetts studies previously referred to a moderate excess of chlorides was not found to be accompanied by a high number of organisms. This fact corresponds with the common observation that vigorous growths are often observed in ponds and lakes far removed from any possible contamination. When the excess of chlorides exceeded 2.5 parts per million larger growths were more frequent, which would tend to show that any relation that exists between microscopic life and excess of chlorides depends upon a relatively large mass of fertilizing elements received from adventitious

sources. Chlorides are a good index of this mass, inasmuch as they remain constant in quantity, for they are not used for plant growth, as are nitrogen compounds, for instance.

UTILIZATION OF ORGANIC SUBSTANCES

General Nature of Organic Food. — Organic matter that is directly or indirectly available as food for microscopic organisms exists in water in both living and inert forms. The former are made up of the cells of bacteria and the plankton, the latter of a great variety of organic compounds found in dead plant and animal cells, products of decomposition and excretions of aquatic animals. All of these contain carbon and nearly all nitrogen, the two most needed elements for cell growth and energy.

Organic Carbon. — We lack means of measurement whereby we can obtain an accurate expression of the amount of different chemical compounds of organic carbon. The total may be obtained and by the use of standard factors an approximation of certain forms of carbon is possible. Thus it may be assumed that proteins contain 53 per cent of carbon, fats 75 per cent, and carbohydrates 45 per cent. The term "total carbon" includes "plankton carbon," that found in the plankton, other organisms and in particulate matter that can be removed by the centrifuge, and "dissolved carbon" which is that in substances in true solution, in a colloidal state, and in organisms too small to be removed by the centrifuge.

Examples of carbon values for lake waters are to be found in the work of Birge and Juday. The plankton carbon of Lake Mendota, Wisconsin, was found to average about 1000 mg. per cubic meter of water, that of plankton nitrogen averaged about 140 mg. per cubic meter. Dissolved carbon greatly exceeded the plankton figures, as did dissolved nitrogen, indicating a large reserve store of food substances. Dissolved carbon and nitrogen values are given in Table 42.

TABLE 42

DISSOLVED ORGANIC CARBON AND ORGANIC NITROGEN IN LAKE WATER
Milligrams per cubic meter of water

		Carbon			Nitrogen		
		Min.	Max.	Mean	Min.	Max.	Mean
Lake Mendota.	14 cases	4000	7,950	5800	289	559	393
Other lakes.	14 cases	3020	13,220	6660	143	750	400

Organic Nitrogen. — Organic nitrogen, on the other hand, may be measured in total amount or in the form of numerous specific compounds that have not been oxidized to ammonia. The mineral compounds of nitrogen likewise are capable of exact determination.

So it is that nitrogen values serve better than others to judge the fertility, active and latent, of natural waters toward plankton growths. Nitrogen in its various forms occurs in widely different amounts in different waters and the amounts fluctuate with the seasons, the pollution and the abundance of plankton life. Other elements necessary for cell life, as hydrogen, oxygen and carbon are in more constant supply than is nitrogen and come from nearly inexhaustible sources, from the water, the air and the carbon dioxide of plant decay.

Forms and Sources of Organic Nitrogen. — Organic nitrogen in lakes and ponds, on the basis of its physical state, may be divided into that in suspension and that in solution, the latter including colloidal solution. Suspended organic nitrogen is contained in complex compounds that are found in bacterial cells, the plankton, larvæ, and floating particles of organic débris. It is sometimes spoken of as plankton nitrogen because it represents floating material and can be removed by filtering or centrifuging the water. In general, these suspended sources contain the most highly organized compounds of nitrogen and carbon found in the plant and animal kingdoms. In many cases the compounds are in the form of living material, as pointed out above.

Soluble Organic Nitrogen. — Soluble organic nitrogen, which term may be extended to include the nitrogen in certain substances in a colloidal state, originates principally from the action of saprophytic bacteria upon inert, insoluble nitrogenous material and from extraction of solid organic particles suspended in the water or precipitated upon the bottom. To a lesser extent excretions of aquatic animal life contribute soluble organic nitrogen. The chemical structure of these soluble compounds is exceedingly varied and their identification in the amounts in which they occur calls for elaborate procedures many of which are comparatively new to chemical science.

A knowledge of these soluble products in water is destined to throw great light upon the factors that control the coming and going of plankton growths in water, especially upon the chemical conditions that surround the manifestation of microscopic life. The amounts in which these soluble compounds of nitrogen occur must not only be studied; information must be forthcoming as to their comparative values for nutrition of plant and animal cells. For example, it has long been known that inorganic nitrogen compounds stimulate plant growth but there is also evidence that amino acids in water greatly increase

the available pabulum. They are indispensable for the nourishment of aquatic animal life.

The presence of amino acids in Wisconsin lake waters has been established by the work of Peterson, Fred, and Domogalla. These authors worked with very large samples of water and checked the accuracy of their determinations in various ways. The quantities of amino acids found by them are given in Table 43. The samples were not taken at the same time, but show that appreciable quantities of these acids occurred in all the lakes examined. The bottom water of Lake Mendota at a depth of 65 feet contained more amino acids than the top water. No correlation was attempted between these single determinations and the plankton life that existed in any of the lakes.

TABLE 43
QUANTITY OF CERTAIN AMINO ACIDS FOUND IN LAKE WATERS
Stated in Milligrams per Cubic Meter

Amino Acid	Method	Mendota Surface, June 18, 1924	Mendota Bottom, June 25, 1924	Devil's Lake, Oct. 10, 1923	Green Lake, July 18, 1923	Lake Michigan, Feb. 28, 1924	Turtle Lake, Jan. 18, 1924
Tryptophane.	Fürth and Nobel.	10.1	13.1	12.2	14.2	5.5	8.6
Tryptophane.	Folin and Looney.	11.0	14.6	12.9	16.4	7.8	10.6
Tryptophane.	May and Rose.	9.9	12.2	.	16.1	6.1
Tyrosine.	Folin and Looney.	10.4	12.5	17.6	9.6	8.3	16.7
Histidine.	Koessler and Hanke.	5.7	10.2	14.8	19.2	6.7	22.7
Cystine.	Folin and Looney.	1.5	6.1	3.3	4.4	2.1	7.5
Total organic nitrogen.....		320	357	177	310	143	487

The same authors attempted a separation of the total soluble nitrogen, organic and inorganic, in Lake Mendota water into its components and succeeded in determining about 90 per cent of the total. The results are given in Table 44.

It will be noted that the plankton nitrogen was much higher in the surface sample and that all forms of soluble nitrogen were higher in the bottom sample. The zone of stagnation is the great storehouse in which all forms of nitrogen accumulate, to be later dispersed through the whole body of water and to stimulate production of plankton

nitrogen. The total of soluble organic forms exceeded that of the inorganic forms in the surface water; in the bottom water the ammonia nitrogen was almost as great as the sum of all the organic forms of nitrogen determined.

TABLE 44
FORMS OF NITROGEN IN LAKE MENDOTA WATER
Stated in Milligrams per Cubic Meter

No.		Surface, June 18, 1924		Bottom, June 25, 1924	
I	Plankton nitrogen.....		92.4		44.9
II	Soluble nitrogen..... (organic and inorganic)		515.6		766.9
	1. Free ammonia nitrogen.....	96.0		280.0	
	2. Residual ammonia nitrogen*.....	16.0		20.0	
	3. Nitrite nitrogen.....	10.0		17.0	
	4. Nitrate nitrogen.....	69.4		92.6	
	5. Free amino nitrogen.....	54.0		81.0	
	6. Peptide nitrogen.....	135.0		140.4	
	7. $\frac{1}{2}$ of tryptophane nitrogen.....	5.3		7.0	
	8. $\frac{2}{3}$ of histidine nitrogen.....	3.8		6.8	
	9. $\frac{2}{3}$ of arginine nitrogen.....	31.1		34.7	
	10. Amide nitrogen.....	12.4		19.3	
	11. Purine nitrogen.....	8.4		9.5	
	12. Amine nitrogen.....	14.2		16.0	
III	Forms of soluble nitrogen determined, total.....		455.6		724.3
IV	Undetermined soluble nitrogen.....		60.0		42.9

* Residual ammonia is that small quantity which remains after distillation of the free ammonia and was obtained by Folin's aspiration method.

Availability of Various Forms of Nitrogen. — Plankton nitrogen is used by "food consuming" organisms. Some protozoa, particularly the flagellated and ciliated species, are bacteria eaters, as are some of the rotifera. The latter also consume algæ and various bacteria eaters among the protozoa. The food of the crustacea is a matter for dispute; it is probable that both algæ and protozoa are used and possible also that organic detritus may be utilized. Ciliated protozoa ingest considerable detritus but the extent to which it serves as a nutrient is not known. Bryozoa and freshwater sponges require plankton nitrogen for their food; they soon die in clean water devoid of plankton life.

Soluble forms of nitrogen are the only ones available for the needs of the synthetic or "food producing" organisms that make up the phytoplankton. The inorganic form, nitrate, is directly available; other mineral forms become so by oxidation.

In addition to mineral substances obtained by assimilation it has been found that the diatoms, as well as the green algæ, may absorb carbonaceous and nitrogenous organic material from the surrounding medium. Hence they must derive their food from two distinct sources.

Of the soluble organic nitrogen compounds that have been shown to exist in lake waters the amino acids are available as food for at least some genera of the phytoplankton and also for aquatic animals. Living organisms may possibly absorb other nitrogen compounds, such as native proteins, amines and amides, but little is known about the nutritive value of these substances to plankton life. There is chemical evidence, however, that the dissolved proteins have a food value equivalent to that of plankton proteids.

Birge and Juday have brought out the fact that the percentage distribution of different forms of organic nitrogen is practically the same in the plankton nitrogen and in the dissolved nitrogen. Their figures are given in Table 45.

TABLE 45
PERCENTAGE DISTRIBUTION OF FORMS OF ORGANIC NITROGEN

	Lake Mendota		12 Other Lakes
	Plankton Nitrogen	Dissolved Nitrogen	Dissolved Nitrogen
Free Amino Nitrogen	17.3%	20.3%	17.2%
Peptide Nitrogen.....	41.7	39.3	38.7
Non-amino Nitrogen.....	41.0	40.4	44.1
Total.....	100.0	100.0	100.0

Quantitative Changes in Nitrogen Content. — The various forms of nitrogen in a lake or reservoir, like the plankton population, are subject to changes, some of which are marked and seasonal, others slight, occasional, and irregular in appearance. The changes among the various forms may be in the same or opposite direction, depending upon relations that normally exist. No two years are likely to show the same variations.

Plankton nitrogen reaches its peak of production in most lakes in the spring and fall, periods corresponding to renewed supply of available food and to its wide dissemination through the vertical depth. Minor peaks appear through the summer as a result of favorable conditions of temperature and light, or because of changes in the inflowing or outflowing water.

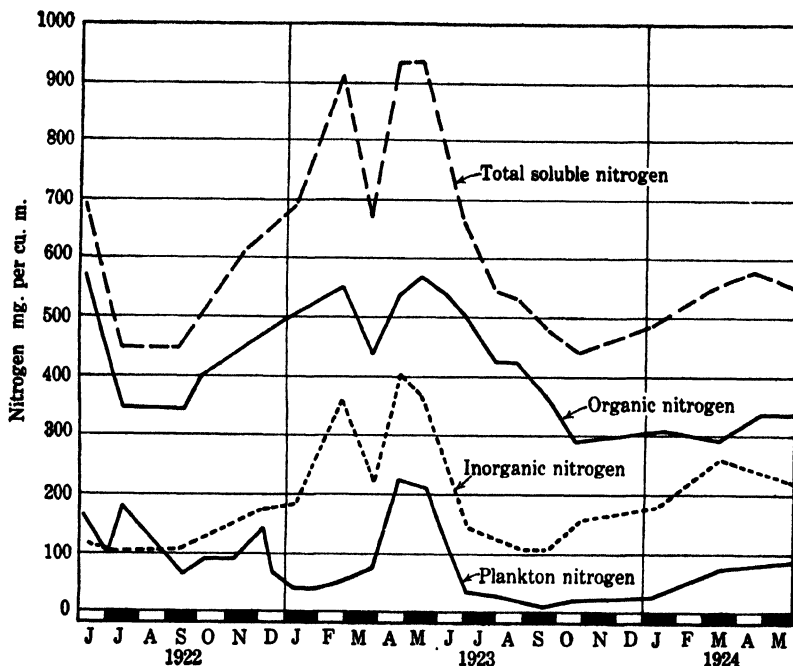


FIG. 60. — Forms of Nitrogen in Surface Water of Lake Mendota.
After Domogalla, Juday, and Peterson.

Other forms of nitrogen than that in the plankton show a variety of relations with plankton nitrogen and with each other. Some of these have been brought out by the studies of Domogalla, Juday, and Peterson on Wisconsin lakes. Figures 60 to 63, constructed from their tabulated monthly results, give a most interesting picture of quantitative changes in nitrogen over a period of two years for both top and bottom water in Lake Mendota. The curves show strikingly different values throughout the course of a year and afford an example of the lack of close agreement in values that may occur for two consecutive years. Some of the conclusions that may be drawn from the work of these authors are given below.

Plankton Nitrogen. — This form of nitrogen was contributed by all insol-

uble material capable of separation by centrifuging, such as bacteria, algæ, protozoa, crustacea and insect larvæ. About 3.5 per cent of the dried material was nitrogen. At all times, as shown by Figs. 60 and 61, there was a large excess of total soluble nitrogen over plankton nitrogen, from three to twenty times as much. The soluble organic nitrogen was also greater than the plankton nitrogen, and usually the soluble

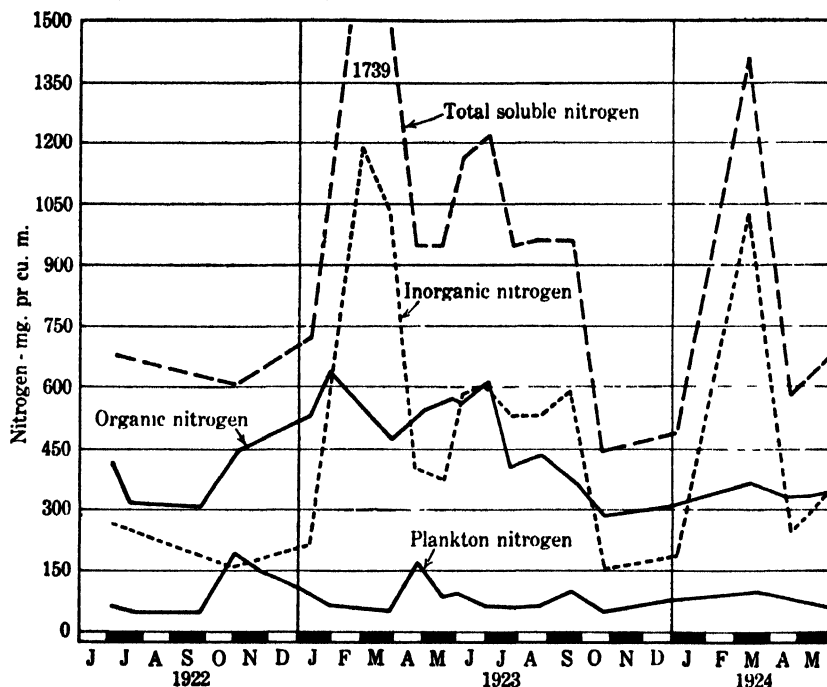


FIG. 61. — Forms of Nitrogen in Bottom Water of Lake Mendota.

After Domogalla, Juday, and Peterson.

inorganic nitrogen was also greater. The curves show spring and fall pulses of plankton growth with less violent fluctuations in the bottom water. In general, plankton nitrogen was higher in the surface water during the summer and lower during the winter, the extreme values in 1923 being 242 mg. per cubic meter (0.242 p.p.m.) in June and 39 mg. per cubic meter (.039 p.p.m.) in February.

Soluble Nitrogen. — This comprises organic compounds and also ammonia, nitrites and nitrates. The estimated proportion of different forms of nitrogen in the soluble nitrogen was as follows:

Ammonia, nitrites, and nitrates	= 25 to 50%
Free amino nitrogen	= 5 to 15%
Peptide nitrogen	= 15 to 35%
Non-amino nitrogen	= 20 to 40%

Soluble nitrogen decreased as plankton nitrogen increased, principally due to consumption by the phytoplankton. Most of the soluble nitrogen is formed in the bottom water and there the most pronounced changes occurred. The seasonal overturns tended to equalize the surface and bottom amounts. Mineral nitrogen increased enormously during the late winter months, in one case as much as 200 per cent in one week, the increases being first noted in the bottom water and finally appearing at the surface. Between January 7 and March 12, 1924, the mineral nitrogen in the bottom water increased from 177 mg. per cubic meter (0.177 p.p.m.) to 1035 mg. (1.035 p.p.m.). The lowest concentration at the surface occurred in the late summer, when organisms

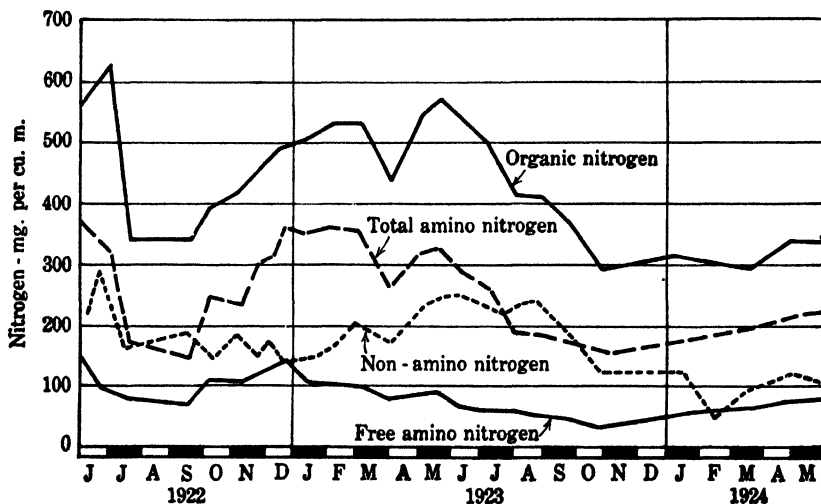


FIG. 62. — Forms of Organic Nitrogen in Surface Water of Lake Mendota.
After Domogalla, Juday, and Peterson.

were still consuming considerable amounts and before much was brought upward from the lower zones.

Amino Nitrogen — This form of soluble organic nitrogen, so valuable for nutritional needs, was divided into two components, free amino nitrogen and peptide nitrogen, on the basis of unhydrolyzed and hydrolyzed residues respectively. Figures 62 and 63 show curves for the fluctuation of free amino and total amino nitrogen, the latter being the sum of the free amino and peptide nitrogen. Both increased during the fall and winter and decreased in the spring and summer. They were more constant in quantity than the mineral nitrogen and did not exhibit great differences in concentration in the upper and lower water. The average of all determinations for total amino nitrogen was about

250 mg. per cubic meter (0.25 p.p.m.) for both surface and bottom samples.

Non-amino Nitrogen. — Non-amino nitrogen consists of the non-amino nitrogen of amino acids and small quantities of nitrogen present as amides, amines and purines. It is probably less utilized by aquatic forms of life than the other forms of soluble nitrogen, a fact that is indicated by the general tendency to remain high in amount during the summer and to show a low level in the winter. The values were not greatly different for surface and bottom samples. In 1923 the surface water varied between 134 mg. per cubic meter (0.134 p.p.m.) in October and 251 mg. (0.251 p.p.m.) in June.

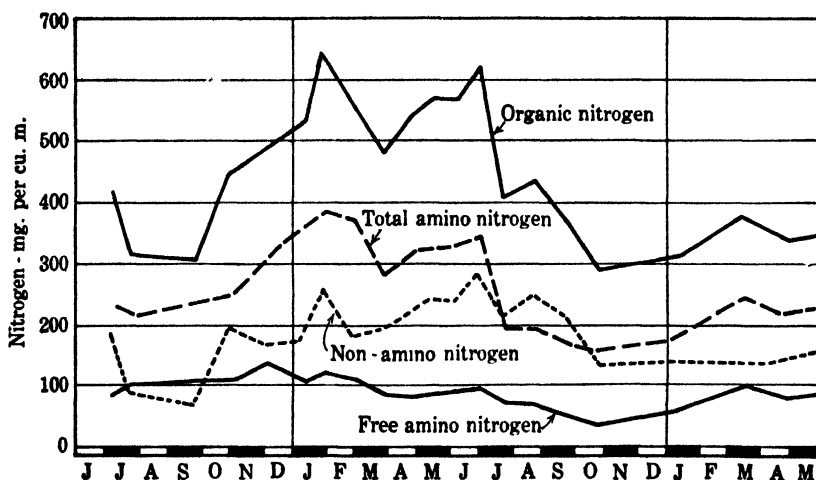


FIG. 63. — Forms of Organic Nitrogen in Bottom Water of Lake Mendota.
After Domogalla, Juday, and Peterson.

Summary of the Wisconsin Work. — The valuable work which has been done upon Wisconsin lakes in connection with the nitrogen content of natural waters contributes much new information and lends support to previously conceived ideas regarding the availability of certain forms of nitrogen for the food of aquatic life. Soluble nitrogen has been quantitatively separated into twelve different forms, which are approximately the same for several lakes. The presence of proteins and amino acids has been established and they have been shown to be at maximum, as are the mineral forms of nitrogen, in the winter, and to reach their low levels in summer. The seasonal variation is an indication that the various forms of soluble nitrogen are part of the pabulum of plant and animal life.

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CHAPTER IX

LIMNOLOGY — BIOLOGICAL CONDITIONS

In the two preceding chapters the physical and chemical constitution of lakes and ponds has been discussed. Methods used in determining the composition of the physical and chemical environment have been given and the effects on plankton activity of the various conditions involved have been outlined. The present chapter deals with the biology of lakes and ponds as pictured by the seasonal distribution, the horizontal and vertical dispersion, and the frequency of occurrence of microscopic organisms. The chapter is confined to these studies because the methods of biological investigation have already been given in Chapters IV and V and because some of the biological aspects of limnology can be better treated in connection with problems such as the storage of water or the control of algæ in water supply systems.

SEASONAL DISTRIBUTION OF MICROSCOPIC ORGANISMS

The microscopic organisms found in water show variations in their seasonal occurrence as great and almost as characteristic as those of land plants. The succession of dandelions, buttercups, and goldenrod in our fields finds its counterpart in the succession of diatoms, green algæ, and blue-green algæ in our lakes and ponds. If one examines the water of a lake continuously for a year some interesting changes in its flora and fauna may be observed. In most lakes of the temperate type the water during the winter contains comparatively few organisms; in the spring various diatoms appear; these disappear in a few weeks and in their place come the green algæ; at the same time, or a little later, the blue-green algæ are found; in the fall both of these vanish and the diatoms reappear; as the lake freezes they in turn disappear. Similar but less characteristic fluctuations take place among the animal forms. These facts are shown graphically in Fig. 64,

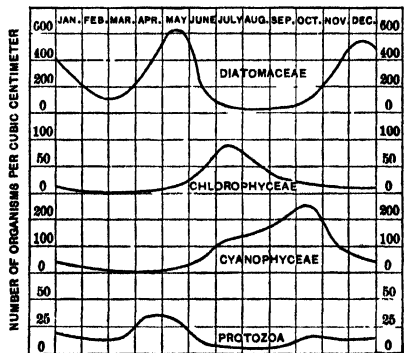


FIG. 64. — Seasonal Distribution of Microscopic Organisms in Lake Cochituate.

which represents the seasonal changes that occur among the more important organisms in Lake Cochituate. The diagram is based on weekly observations extending over a number of years. The seasonal distributions of the diatoms, algæ, and protozoa, are so different that it is best to consider each class by itself.

Diatomaceæ. — In most natural ponds and storage reservoirs diatoms are far more abundant in spring and fall than at other seasons. New growths seldom begin in summer or winter, but the spring and fall growths sometimes linger for a number of weeks into the summer and winter.

The occurrence of diatoms in ponds is greatly influenced by the vertical circulation of the water. They generally appear after the periods of stagnation and during the periods of complete vertical circulation. It has been found that in temperate lakes of the second order, which have well-marked periods of stagnation in summer and in winter, the spring and fall growths of *Asterionella* occur with great regularity and with about equal intensity, while in temperate lakes of the third order, which are stagnant only during the winter, the development of *Asterionella* in the autumn is either small compared with the spring growths or altogether lacking. In deep ponds the spring growths occur earlier and the fall growths considerably later than in shallow ponds, thus again corresponding to the periods of circulation. In lakes of the third order diatoms are sometimes found during the summer after periods of partial stagnation.

Of the many genera of diatoms that are observed in water only those that are truly planktic exhibit the spring and fall maxima. The most important of these are *Asterionella*, *Tabellaria*, *Melosira*, *Synedra*, *Stephanodiscus*, *Cyclotella*, and *Diatoma*. Other genera are more uniformly distributed through the year. All of the seven genera listed above are sometimes, but not often, observed during the same season in the same body of water. As a rule certain ponds have certain diatoms peculiar to them. For example, Lake Cochituate often contains large growths of *Asterionella*, *Tabellaria*, and *Melosira*; other diatoms are to be found, but they are seldom very numerous. Sudbury Reservoir, No. 3 of the Boston Water Works contains *Asterionella*, *Tabellaria*, and *Synedra*, but few *Stephanodiscus* or *Melosira*. In Sudbury Reservoir No. 2 only *Synedra* and *Cyclotella* are found. In the Ashland Reservoir *Cyclotella* usually predominates. Fresh Pond, Cambridge, Mass., is famous for its *Stephanodiscus*, and *Diatoma* is common in the water supply of Lynn, Mass.

The genera that appear in any pond are not the same every year. In Lake Cochituate the spring growth in 1890 consisted of *Asterionella*

and *Tabellaria*; in 1891 of *Asterionella* with a few *Melosira*; in 1892 chiefly of *Melosira*; in 1893 of *Melosira* and *Asterionella*; and in 1894 of *Tabellaria*, *Asterionella*, and *Melosira*. Furthermore, in any season it is seldom that two genera attain their maximum development simultaneously — sometimes one appears first and sometimes another. The most interesting succession of genera that the author has observed occurred in 1892 in Chestnut Hill Reservoir of the Boston Water Works.

The spring growth began in April and continued through July. For three months the total number of diatoms present did not materially change, but during this time six different genera appeared on the scene, culminated one after another, and disappeared. This is shown in Fig. 65.

The explanation of the peculiar seasonal distribution of diatoms involves the answers to many questions. To what extent are diatoms influenced by light, by temperature, by mechanical agitation? To what

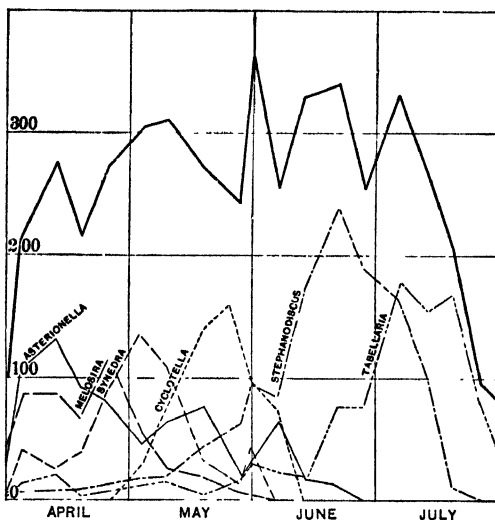


FIG. 65. — Succession of Diatoms in Chestnut Hill Reservoir, 1892.

extent are they dependent upon oxygen or carbonic acid dissolved in water? What sort of mineral matter do they require? These are questions not yet fully answered. Attempts have been made to solve the problems by experiment, but it has been found difficult to control all the necessary conditions in the laboratory.

Relation of Temperature to Diatom Growth. — The optimum temperature for the development of diatoms is not known. Diatom growths have been observed at temperatures ranging from 35° to 75° F. In Lake Cochituate the average temperature of the water at the time of maximum *Asterionella* growths is not far from 50°. In some lakes it is nearer 60°. There are reasons for believing that the optimum temperature for the diatoms is lower than for the green or blue-green algae. This theory is substantiated by a study made by Dr. Frank E. Hale on the growths of diatoms and blue-green algae in the Croton catchment area during 1921. The occurrence of growths of over 1000 units with reference to temperature is shown in Table 46.

The frequency of large growths of diatoms was greatest between 40° and 60° F., while cyanophyceæ predominated in great numbers when the water was warmer. The blue-greens showed a steady increase in frequency of occurrence with rising temperatures.

TABLE 46
RELATION OF TEMPERATURE TO MICROSCOPIC GROWTHS
Number of occurrences of growths of 1000 standard units or over
Croton Reservoirs, 1921
After Hale

Organism	Temperature °F.				
	30-40	40-50	50-60	60-70	70-80
<i>Diatomaceæ:</i>					
Synedra.....	3	0	5	2	1
Melosira.....	1	5	4	1	1
Fragilaria.....	1	0	0	0	0
Asterionella.....	1	8	7	1	0
Tabellaria.....	0	2	0	0	0
Total.....	6	15	16	4	2
<i>Cyanophyceæ:</i>					
Aphanizomenon.....	5*	10	4	7	4
Anabæna.....	0	5*	7*	9	12
Celosphærium.....	0	0	0	0	1
Mixture.....	2*	2*	4	5	13
Total.....	7	17	15	21	30

* Persistence, not start of growth.

Relation of Light to Diatom Growth. — It is known that diatoms are very sensitive to light. They do not grow in the dark nor in bright sunlight. Experiments made by the author in which diatoms were allowed to grow in bottles at various depths below the surface have shown that their growth is nearly proportional to the intensity of the light. This is illustrated by Fig. 66. It will be noticed that near the surface,* where the light was strong, they multiplied rapidly, but below the surface the rate of multiplication was much slower, and at a certain depth no multi-

* The growth at the depth of 6 inches was greater than at the immediate surface, where the direct sunlight was too strong.

plication took place. This depth-limit of growth varied according to the color and transparency of the water, being greatest in the water having the least color. In one reservoir, where the color was 86 p.p.m., the limit of growth was 8 ft.; in another, where the color was 60, it was 12 ft.; and in a third, with a color of 29, it was 15 ft. No observations were made in colorless waters, but the limit of growth in them is probably as great as 25 or 50 ft., and perhaps even more.

The specific gravity of diatoms plays an important part in their seasonal distribution. In absolutely quiet water most diatoms sink to the bottom, but very slight vertical currents are sufficient to keep them in suspension. A few forms appear to have a slight power of buoyancy, and some genera are somewhat motile. Diatoms also liberate during growth oxygen gas which may give them buoyancy.

Diatoms are said to be positively heliotactic, that is, they tend to move toward the sunlight. In some of the motile forms this power is quite strong. In most of the plankton genera it is weak; they will not move upward toward the light through any great depth of water. It is possible, however, that the power of heliotaxis varies with the intensity of the light, but experimental evidence on this point is lacking.

Relation of Food Materials to Diatom Growth.—Diatoms require oxygen for their best development. Experiments have shown that they will not multiply in a jar in which a thin layer of oil covers the surface of the water; that in cultures in jars of various shapes, the one that has the least depth of water and the greatest amount of surface exposed to the air will show the greatest multiplication; that in bottles exposed at the same depth beneath the surface of a reservoir, those with bolting cloth tied over the mouth will show a greater development of diatoms than those tightly stoppered.

The nature of the food material of diatoms is not well known (see Chapter VIII). Observations seem to show that they require carbon, nitrogen in the form of nitrates or free ammonia (perhaps both), silica, and more or less mineral matter, such as the salts of magnesium, calcium, iron, and manganese, but the amounts of these various substances required have not been determined.

The facts at hand enable one to formulate a theory for the explana-

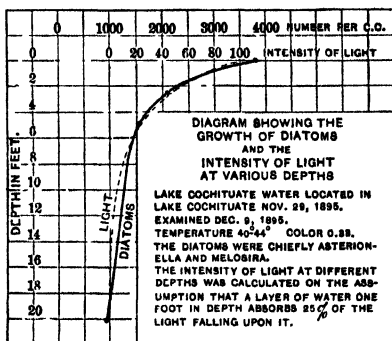


FIG. 66. — Relation of Light to Diatom Growth.

tion of the occurrence of maximum growths of diatoms after the periods of stagnation and during the periods of circulation.

During the periods of stagnation the lower stratum of water in a deep lake undergoes certain changes that are very pronounced if the bottom of the lake holds any accumulation of organic matter. The organic matter decays, oxygen becomes exhausted, decomposition proceeds under the action of anaërobic bacteria, free ammonia increases, and other organic and inorganic substances become dissolved in the water. During the period of circulation this foul water reaches the surface, oxidation takes place, and compounds favorable to the growth of diatoms are carried upward. At the same time the vertical currents carry to the surface the diatoms, or their spores, that have been lying dormant at the bottom, because of darkness or the absence of proper nutriment conditions. Lifted toward the surface, where there is an abundance of light, air, and food, they multiply rapidly. The extent of their development depends upon the amount of food material present, the temperature of the water, and the amount of vertical circulation. If the upper layers become stratified and the surface remains calm for a number of days the diatoms settle back into a region where the light is less intense. If they sink low enough they enter a region where the light is not sufficient for growth, and if they sink below the transition zone succeeding vertical circulation of the upper strata does not affect them. Unable to reach the surface by their own power they fall to the bottom and remain dormant through another period of stagnation.

In small reservoirs that are constantly supplied with water rich in diatom food and that are so shallow that even at the bottom the light is strong enough for diatom development, the seasonal distribution follows somewhat different laws. This is the case in many open reservoirs in which ground water is stored.

Chlorophyceæ. — The grass-green algæ are most abundant in water supplies during the summer. They are seldom found in winter. The curve of their development is more nearly parallel to the curve of water temperature than that of any other class of organisms. The maximum growth occurs usually in July or August, though the development of some genera culminates as early as June and that of others as late as September or even October. The late growths are usually associated with the phenomenon of stagnation.

The optimum temperature for the different genera is not known. It seems probable that most of the common forms are able to grow vigorously between 60° and 80° F. if their food supply is favorable and the light is sufficient. It is possible for some of the green algæ to become accustomed to considerable extremes of heat or cold. *Protococcus*

nivalis is found in the arctic regions, and *Tribonema* has been observed in water at a temperature of 115° F.

Cyanophyceæ. — The seasonal distribution of the cyanophyceæ is similar to that of the chlorophyceæ, but as a rule the maximum growths occur a little later in the season. The blue-green algæ seem to be attuned to a slightly higher temperature than the grass-green organisms and, as shown in Table 46, to a considerably higher temperature than the diatoms. They often show a great increase after a period of hot weather. *Anabæna*, *Clathrocystis*, and *Cœlosphærium* seldom give trouble unless the temperature of the water is above 70° F. This explains why excessive growths of blue-green algæ rarely occur in England where even in summer the surface water seldom reaches so high a temperature.

Aphanizomenon is more tolerant of temperature conditions (see Table 46). It apparently prefers colder water than most of the cyanophyceæ. In some ponds it is present throughout the entire year, even when the surface is frozen. On one occasion it grew under the ice in Laurel Lake, Fitzwilliam, N. H., and became frozen into the ice to such an extent that the ice cutters were alarmed at the green color imparted to the ice. In Lake Cochituate, *Aphanizomenon* reaches its greatest growth in the autumn. This accounts for the maximum of the curve of cyanophyceæ in Fig. 64 occurring in October instead of in August or September.

Schizomycetes and Other Fungi. — These forms have no well-marked periods of seasonal distribution. They are liable to occur at any time of year. Mold hyphæ are occasionally found at the bottom of lakes during the summer, and at the surface under the ice in winter. *Crenothrix* may be found in the stagnant water at the bottom of a deep lake during the summer, and at all depths in the autumn after the overturning of the lower layers of water. *Crenothrix* has been observed during the summer in swamps in company with *Anabæna* and other cyanophyceæ. Attention is again called to the possibility of mistaking the stems of *Anthophysa* for *Crenothrix*.

Protozoa. — The seasonal distribution of the protozoa, taken as a group, is extremely variable and differs considerably in different ponds. No curve can be drawn that will represent all cases. In Lake Cochituate the curve has a major maximum in the spring, a summer minimum lower than that which occurs in the winter, and a minor maximum in the autumn. In Mystic Lake the curve has but one maximum — in the summer. These differences are due to the fact that the group of protozoa is a broad one, and includes organisms that differ widely in their mode of life.

The sarcodina are found at all seasons of the year, but they are most numerous in the plankton in the autumn after the period of summer stagnation. These organisms live upon the ooze on the bottom and sides of ponds and upon twigs and aquatic plants. There they are found most abundantly in the summer. The vertical currents of the autumnal circulation scatter them through the water and cause the maximum number of floating forms to be observed during October and November. There is a minor maximum during the period of spring circulation. Some planktic forms, such as *Actinophrys*, are most abundant in summer.

Of the mastigophora the flagellata, *Euglena*, *Raphidomonas* and *Phacus* are most abundant from June to September; *Trachelomonas* is found at all seasons, but is most common in the fall after the period of summer stagnation; *Mallomonas* is found from April to October, but is usually most abundant in the autumn; *Cryptomonas* occurs in some ponds only in the late fall and winter; *Synura* and *Dinobryon* are generally most numerous in the spring and autumn, but heavy growths have been observed at all seasons; *Uroglenopsis* seems to prefer cold weather, but vigorous growths have been noted in June.

The dino-flagellata, *Glenodinium* and *Peridinium*, are usually most abundant during warm weather, but they are liable to occur at any season. *Ceratium* seldom appears before July and usually disappears before cold weather.

Of the infusoria, most of the ciliated forms prefer warm water; *Codonella* and *Tintinnus* occur after periods of stagnation; *Vorticella* and *Epistylis* are distinctly summer organisms; and *Bursaria* and *Stentor* are also found in summer.

Acineta is most abundant during warm weather.

The protozoa that attain their greatest development in summer are those forms that are closely allied to the vegetable kingdom, and that are perhaps more properly classed with the algæ: namely, the dino-flagellata and some of the flagellata that are rich in chlorophyll. A few genera that occur most abundantly in the spring and fall have a brownish-green color like that of the diatoms, which also have spring and fall maxima. The ciliata that live upon decaying organic matter are attuned to a comparatively high temperature — about 75° F. This has been demonstrated by experiment and corresponds with the time of their observed maximum. The protozoa that exhibit a strictly animal mode of nutrition are most abundant during those seasons when there is plenty of food material in the shape of minute organisms or finely divided particles of organic matter. This partially explains why growths are sometimes present in the winter when bacteria are numerous or after

periods of stagnation when particles of organic matter from the bottom have been scattered through the water.

Rotifera. — Rotifera are found at all seasons of the year, but are most numerous between June and November. In many ponds the maximum occurs in the autumn. Some genera are perennial, others are periodic in their occurrence. *Anuræa* and *Polyarthra* are found throughout the year, but their number rises and falls at intervals corresponding to the hatching season. *Conochilus* is often abundant in June, *Asplanchna* in July and August, and *Synchaeta* in August and September. The littoral rotifera are most numerous during the summer.

The rotifera feed upon the smaller algæ, and their seasonal distribution is largely influenced by the amount of this food supply. The reactions of the rotifera to light, temperature, etc., are not well known.

Crustacea. — The number of crustacea found at different seasons varies greatly in different bodies of water. It is influenced largely by the genera that are present. Different genera vary considerably in their seasonal distribution. Some are found at all seasons, while others occur only at certain times. The perennial forms may have several maxima during the year, corresponding to the hatching of different broods. As a rule crustacea are most numerous in the spring, but minor maxima may occur during the summer and autumn and rarely in the winter.

Temperature, food supply, and competition are the chief factors that influence the seasonal distribution of the crustacea.

For a full discussion of the seasonal distribution of the crustacea the reader is referred to Dr. Birge's studies of the crustacea of Lake Mendota. The organisms are given scant attention in this book because they have but little direct significance in public water supplies.

Annual Variation in Seasonal Distribution. — Microscopic growths do not follow the calendar but fluctuate with the seasonal changes in the conditions of existence. We have seen that the environmental factors such as temperature, light, water movement, and food supply, combine in different ways during the seasons to produce seasonal maxima and minima of plankton crops. Seasons vary from year to year. They are similar but not the same. As a result the seasonal occurrence and magnitude of plankton growths also vary from year to year. The general seasonal trend is similar but the actual volume of organisms present in the same body of water during any particular season varies in different years. This irregularity of the seasonal occurrence of microscopic organisms may be seen from Fig. 67, which shows the changes in growth of diatoms and blue-green algæ that took place in the water of Lake Cochituate during a period of five years. *

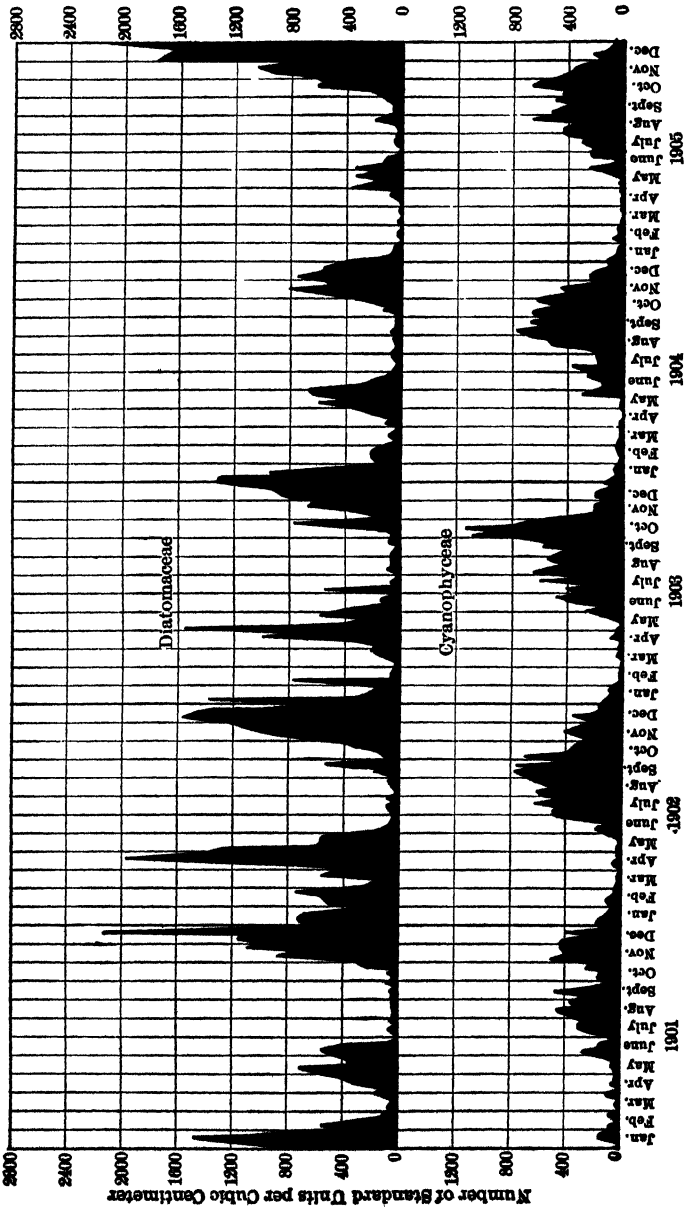


FIG. 67. — Seasonal and Annual Distribution of Diatoms and Blue-green Algae in Lake Cochituate, 1901 to 1905.

HORIZONTAL AND VERTICAL DISTRIBUTION OF MICROSCOPIC ORGANISMS

The plants and animals that inhabit lakes and ponds may be classified according to their habitat, as *littoral*, *benthic*, or *limnetic* organisms.

The *littoral* and *benthic* organisms include all those forms that are attached to the shore and bottom, or to plants growing on the shore and bottom, besides a host of others that, though free-swimming are almost invariably associated with the attached forms.

The *limnetic*, or *pelagic*, organisms are those that make their home in the open water. They float or swim freely and are drifted about by every current. Collectively they make up the greater part of the microscopic organisms and include almost all the troublesome odor-producing organisms of water supplies. In the open water, one often finds, too, some of the littoral forms that have come detached from the shore and scattered through the water by the currents, or that are parasitically attached to some of the limnetic forms. Then there are organisms that may be said to be *facultative limnetic forms*, because they thrive in the open water as well as on the shore. The true limnetic forms, however, are the most important in water supplies, and a study of their horizontal and vertical distribution is therefore of value.

Horizontal Distribution. — The horizontal distribution of the limnetic organisms is usually quite uniform within any limited area, but through the entire body of a lake the number of organisms may show considerable variation. This is quite noticeable in long, narrow reservoirs in which streams enter at one end and discharge at the other. In such reservoirs the organisms are generally most numerous at the lower end. If, however, the water in the incoming stream contains many organisms the numbers may be higher at the upper end, diminishing gradually as the water of the stream becomes mixed with that of the reservoir. Sometimes the mixing takes place slowly and the influent water passes as a current far into the reservoir. This tends to distribute the organisms in streaks. In lakes with uneven margins the horizontal distribution may vary greatly, and the number of organisms found in coves may be quite different from the number found in the open water. The horizontal distribution of diatoms is influenced to some extent by the depth of the lake. There exists for example in Massachusetts a lake covering about 250 acres. Near one side of it there is a deep hole, that has an area of about five acres. Here stagnation phenomena are very pronounced. When growths of diatoms occur in the spring and fall the numbers are much higher in the vicinity of this deep hole than elsewhere in the lake.

Areas of shallow flowage exert a marked effect on the horizontal distribution of the microscopic organisms.

The wind also has a great influence, and in many bodies of water wind is the controlling factor. The organisms, particularly the cyanophyceæ, are driven in the direction of the wind and accumulate toward the windward shore.

Undertow currents also play an important part in the horizontal distribution of organisms. Algæ that have developed within the transition zone may by a sudden increase in wind movement be carried into the circulating waters near the surface.

Vertical Distribution. — The laws that govern the vertical distribution of microscopic organisms are more complicated than those determining their horizontal dispersion. The latter are less important than the former because they affect the organisms only mechanically, while the former affect them vitally.

In a lake of the second order the factors determining plankton growth vary at different depths and at different seasons. In the summer, for example, the conditions above the transition zone are very different from those below it. Near the surface the water is warm, the light strong, and oxygen abundant. Vertical currents are active and carbonic acid is present early in the season. Near the bottom the water is cold and the light weak. The oxygen may be exhausted, and the water is perfectly quiet. Under these circumstances chlorophyll-bearing organisms naturally thrive best above the transition zone. They seldom develop below it. Often they are found concentrated within the transition zone itself.

It has been shown that under experimental conditions the development of diatoms is greatest near the surface and that it decreases downward as the light decreases. In nature, however, it cannot be expected that the number of diatoms in the different layers of water will follow this law closely, because the diatoms are heavy and constantly tend to sink, and because the water above the transition zone is more or less stirred up. One would expect rather to find a uniform vertical distribution above the transition zone, and below it a rapid decrease in the number of organisms. Such a distribution is common. The following instances of the vertical distribution of *Asterionella* and *Tabellaria* in Lake Cochituate may be cited in illustration; in both cases the transition zone was located between 20 and 30 ft.

This manner of distribution is most common during periods of rapid development, when a gentle breeze is stirring. In very calm weather and during periods of declining growth diatoms sink rapidly, and at such times they may be found most numerous at the transition zone or

at the bottom. During periods of complete vertical circulation the vertical distribution may be quite uniform from top to bottom. The diatoms found at the bottom of a deep lake are usually less vigorous than those near the surface.

TABLE 47
VERTICAL DISTRIBUTION OF ASTERIONELLA AND TABELLARIA IN
LAKE COCHITUATE

Depth in Feet	Numbers per cc.	
	Asterionella. May 7, 1891	Tabellaria. May 24, 1890
Surface	3752	1886
10 ft.	3736	1448
20 "	3716	1396
25 "	...	484
30 "	1784	298
40 "	456
50 "	536
60 "	178	96

The chlorophyceæ and cyanophyceæ are much lighter in weight than the diatoms, and some of them contain oil globules and bubbles of gas that increase their buoyancy. The forces tending to keep them near the surface are greater, therefore, than is the case with the diatoms. The grass-green and blue-green algæ are seldom found below the transition zone, and their distribution varies at different depths within the circulation zone. The cyanophyceæ collect near the surface. In quiet waters they often form unsightly and ill-smelling scums or water bloom. Occasional exceptions to the general vertical distribution are observed. Microcystis, for example, is usually more abundant in Lake Cochituate just below the transition zone than it is at the surface. On July 31, 1895, the number of standard units of Microcystis at different depths was as follows: Surface, 94; 30 ft., 342; 60 ft., 140. Hale reports the following distribution of diatoms and blue-greens in the Croton reservoirs in 1921 as indicated by surface and bottom samples (Table 48).

It is interesting to note that a sudden wind may affect the vertical distribution of the cyanophyceæ and the diatomaceæ in opposite ways. It may tend to decrease the number of blue-green algæ at the surface by preventing the formation of scums, while it increases the number of diatoms by keeping them from sinking.

TABLE 48
 SURFACE AND BOTTOM GROWTHS OF DIATOMS AND BLUE-GREEN ALGÆ
 Number of occurrences of growths of 1000 standard units or over
 Croton Reservoirs, 1921
After Hale

	Surface	Bottom
<i>Diatomaceæ:</i>		
First six months.....	14	8
Second six months.....	5	4
Total.....	19	12
<i>Cyanophyceæ:</i>		
First six months.....	3	1
Second six months.....	42	22
Total.....	45	23
Total:		
First six months.....	18*	9
Second six months.....	47	26
Total.....	65	35

* One occurrence of Dinobryon, a protozoön.

The protozoa, as a class, seek the upper strata of water. *Euglena* sometimes forms a scum upon the surface. In winter *Uroglenopsis*, *Synura*, etc., are often most numerous just beneath the ice. The dino-flagellata are distinctly surface forms. Some of the protozoa seem to avoid direct sunlight and keep away from the upper surface of the water, though they may be very abundant at a depth of one or two feet. As elsewhere pointed out, many of these organisms contain chlorophyll and perhaps should be classed as algæ. The ciliata and other holozoic protozoa which have a distinctly animal mode of nutrition are more irregularly distributed through the vertical. The sarcodina are most abundant near the bottom.

Flotation of the Plankton. — Some of the microscopic organisms are heavier than water, some are lighter, and some have about the same specific gravity. Various means are used by the heavier organisms to float themselves.

1. Some secrete a gelatinous watery envelope that is lighter than water.

2. Some form vacuoles.

3. Some produce substances lighter than water, such as:

- a. Gas confined in the upper parts of the bodies or in special holders.
- b. Oily or fatty substances.

4. Some expand their surface area and thus increase the surface friction with the water. This is accomplished in several ways:

- a. By the enlargement of the entire surface.
- b. By the formation of grooves, or markings, as in some of the diatoms.
- c. By the attachment of many cells to form a filament and by the development of long needle-like forms.
- d. By the formation of special swimming attachments, as the cilia and flagella of protozoa, and the antennæ and legs of crustacea.
- e. By the formation of colonies of organisms of considerable size.

Concentration of Organisms in the Transition Zone. — At times some of the microscopic organisms are more numerous in the transition zone than elsewhere in the vertical. An interesting illustration of this occurred in Lake Cochituate in the summer of 1896. *Mallomonas* is not ordinarily abundant in this lake, but on June 24 it suddenly appeared just below the upper boundary of the transition zone. At mid-depth (30 ft.) there were 116 of these organisms per cc., at the bottom 42 per cc. but at the surface there were none. They developed rapidly, and on August 4 there were 3640 at mid-depth. The growth continued until September, and during this time the largest number observed at the bottom was 276 per cc., while above the transition zone scarcely an individual was found. On July 17 the vertical distribution was as follows:

TABLE 49

VERTICAL DISTRIBUTION OF *MALLOMONAS* IN LAKE COCHITUATE, JULY 17, 1896

Depth, Ft.	Number per cc.	Temperature °F.
Surface	0	77.3
10	0	75.2
15	2	62.0
20	1454	47.7
25	794	43.7
30	548	43.2
40	112	42.5
50	88	41.4
60	64	40.8

Synura and other organisms have shown a similar vertical distribution and the phenomenon is probably more common than we've used to think. Whether this concentration at the transition zone is due to food material, to light, or to temperature is not definitely known. *Mallomonas* is motile and known to be positively heliotactic. In the winter members of this genus are often numerous under the ice. It is possible that they have a low temperature attunement, and that in the instance above cited they collected as near the surface as their temperature attunement would permit. This accords with the fact that they are most numerous in the spring and fall. It is possible that the dissolved gases are a factor in the problem and also the increased density and viscosity of the water at lower temperatures. Supersaturation of the water with oxygen at the transition zone has already been alluded to.

The organisms most frequently found concentrated at the transition zone, partake of the animal nature. Presumably they depend, in part at least, for food upon other organisms, as for example, bacteria, and upon organic detritus. It is possible that in the process of sedimentation in a lake these substances are temporarily checked in their fall by reason of the greater density and viscosity of the colder water at the transition zone, and that the protozoa, some of which are bacteria eaters, congregate there to devour them; while the crustacea congregate there to devour the protozoa and the detritus.

This does not apply to the chlorophyllaceous organisms, such as *Aphanizomenon*, *Mallomonas*, *Synura*, and *Dinobryon*, which are often found concentrated in the transition zone.

Rotifera and crustacea are often numerous above the transition zone, but are commonly more abundant in or below it. Apparently their food supply is a controlling factor. During the winter they are sometimes numerous at the bottom. Different genera react differently to light, and heat. Some of them show a slight daily migration toward the surface at night, and away from the surface in the daytime.

The schizomycetes are usually more abundant at the bottom of a pond than at the surface. Mold hyphæ are often numerous in winter just under the surface of the ice.

Adaptation of Organisms to Changed Viscosity of Water. — The viscosity of water changes greatly with variations in temperature. At 25° C. (77° F.) the viscosity of water is only one-half of that at 0° C. (32° F.), consequently the tendency of organisms to sink at 25° is about twice that at 0° C. Concomitant with the decrease in viscosity is a reduction in density. Unless the organisms can adapt themselves to these changes and in some way increase their buoyancy during warm weather they will sink to a colder stratum of water and perhaps even

to the bottom. Possibly the slight diurnal changes in the temperature of the upper water strata may be a factor in the vertical migration of certain crustacea, the organisms rising to the surface as the water cools at night and sinking back as the sun warms the water.

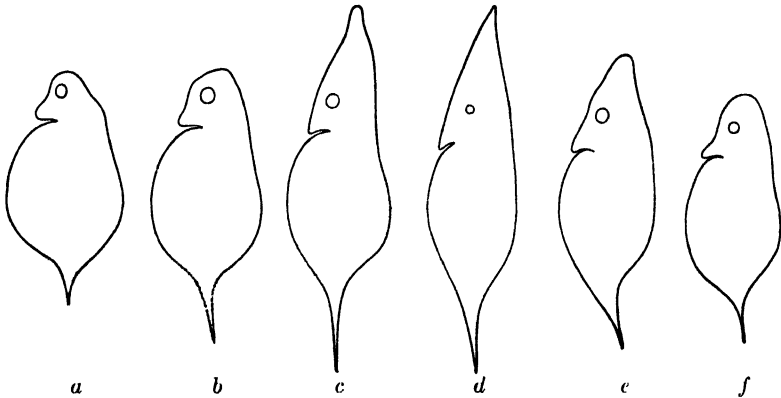


FIG. 68. — Seasonal Change in Shape of *Hyalodaphnia*. *a*, *b*, and *f* are winter forms; *c*, *d*, and *e* are summer forms.

Dr. C. Wesenberg-Lund claims that certain organisms adapt themselves to changes in viscosity, by expanding during warm weather, thus increasing the surface exposed to the water, or by changing their shape or the location of their center of gravity. This theory is interesting, but it has not been fully demonstrated. *Daphnia hyalina* is round-

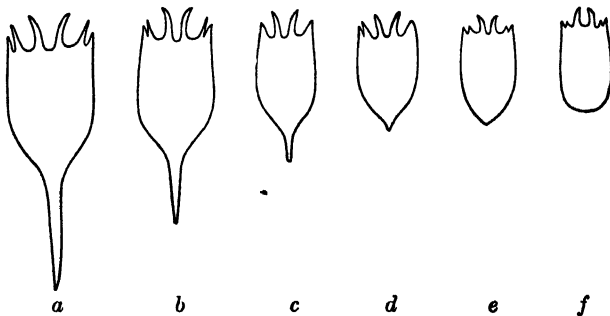


FIG. 69. — Seasonal Changes in Shape of *Anuræa*. *a*, *b*, and *c* are summer forms; *d*, *e*, and *f* are winter forms.

headed during the winter but point-headed during the summer; *Bosmina coregoni* enlarges in summer; *Asplanchna priodonta* becomes elongated; while *Ceratium hirundinella* grows an extra horn that increases its floating power. *Tabellaria* increases the number of cells in its colonies and thus attains greater flotation and doubtless other diatoms

do the same. These changes take place at a temperature of 12° to 16° C. (47.6° to 60.8° F.), that is during May and June, and again in the autumn; the change is not gradual but takes place in the course of two or three weeks.

Wesenberg-Lund has also shown that these variations are regional as well as seasonal. There is a gradual decrease in volume of many well known plankton forms from the south to the north, and in regions where there is the greatest range of temperature there is also the greatest seasonal variation. The low temperature forms of the plankton tend to uniformity, but the high temperature forms vary in different lakes.

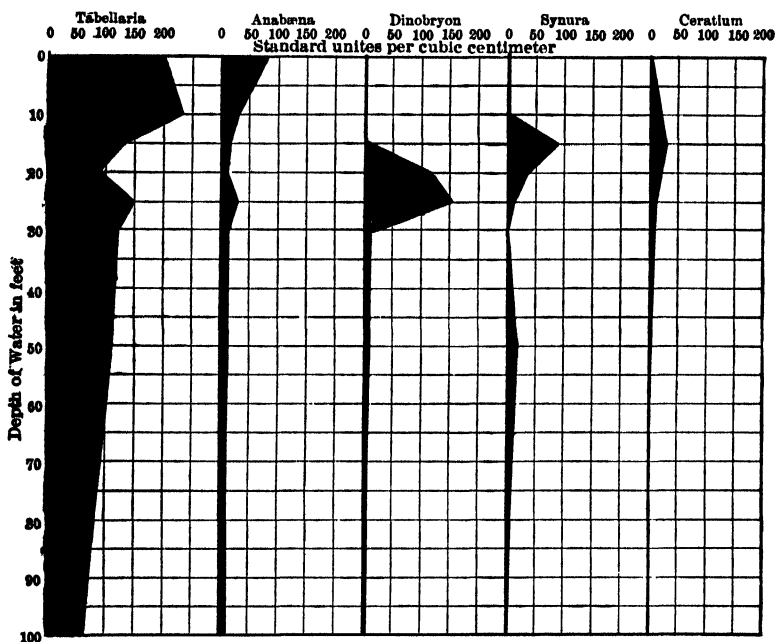


FIG. 70. — Vertical Distribution of Microscopic Organisms in McGregor Lake, near Ottawa, Ontario. July 12, 1911.

Examples of Vertical Distribution. — Figure 70 shows the distribution of certain organisms in McGregor Lake situated in the Province of Ontario a few miles north of Ottawa. Here in July, 1911, it was found that the diatom, *Tabellaria* and the blue-green alga, *Anabæna*, were most abundant near the surface, but that *Dinobryon* and *Synura* were much more abundant in the transition zone. The studies in this lake were of especial interest by reason of its high latitude.*

* The full report by the author was published in the Annual Report of the Provincial Board of Health of Ontario, Canada for the year 1911.

In spite of the tendencies of the organisms to choose their favorite habitat in a body of water, the mechanical effects of winds, currents, gravity, and other factors are so great that in most ponds and reservoirs used for water supply, except in very deep ones, the average number of organisms of all kinds through the year does not vary much at different depths. This is illustrated by the following table:

TABLE 50
VERTICAL DISTRIBUTION OF MICROSCOPIC ORGANISMS. BOSTON WATER WORKS
1890-1896

	Depth	1890	1891	1892	1893	1894	1895	1896
Lake Cochituate	Surface	454	736	523	389	416	355	507
	30 ft.	304	569	528	336	365	373	657
	60 ft.	357	650	626	316	309	353	544
Sudbury Reservoir No. 2	Surface	68	322	268	116	45	61	87
	13 ft.	80	273	256	98	49	56	120
	25 ft.	64	268	229	98	33	47	78
Sudbury Reservoir No. 3	Surface	152	277	514	381	289	621	524
	15 ft.	182	267	523	303	194	543	467
	30 ft.	131	323	481	311	179	485	498
Ashland Reservoir	Surface	50	129	269	112	28	57	94
	20 ft.	38	95	268	84	20	35	108
	40 ft.	25	83	235	66	20	25	106
Hopkinton Reser- voir	Surface					87	105	189
	25 ft.					52	58	118
	50 ft.					72	53	104

For the years 1890 to 1893 the results were given in Number of Organisms per cc. Since Jan. 1, 1893, the results have been given in Number of Standard Units per cc.

The vertical distribution varies at different seasons, as the following table illustrates:

TABLE 51

SEASONAL VARIATION IN MICROSCOPIC ORGANISMS AT THE SURFACE, MID-DEPTH, AND BOTTOM. RESERVOIRS OF THE BOSTON WATER WORKS, 1895

Standard Units per cc.

	Depth	January	February	March	April	May	June	July	August	September	October	November	December	Mean
Lake Cochituate	Surface	255	34	10	97	188	437	480	248	137	450	1159	762	355
	30 ft.	407	21	23	109	149	188	539	329	193	400	1199	921	373
	60 ft.	422	232	55	101	133	188	503	290	53	252	1198	808	353
Sudbury Reservoir No. 2	Surface	6	8	6	49	56	109	163	152	82	72	15	18	61
	13 ft.	6	7	18	25	59	76	195	108	93	53	14	17	56
	25 ft.	4	7	17	22	47	63	160	88	74	49	22	9	47
Sudbury Reservoir No. 3	Surface	13	3	14	62	375	787	1197	1675	1778	1227	206	53	621
	15 ft.	18	1	14	46	260	768	1072	1134	1813	1161	253	34	543
	30 ft.	47	4	13	57	235	597	633	1146	1487	1342	222	37	485
Aahland Reservoir	Surface	78	74	10	27	79	76	123	75	30	45	40	22	57
	20 ft.	18	19	5	15	37	47	43	78	29	55	37	19	35
	40 ft.	13	19	18	12	21	41	48	38	7	33	21	26	25
Hopkinton Reservoir	Surface	41	50	36	64	91	193	203	91	243	186	41	13	105
	25 ft.	28	10	4	57	42	61	46	65	130	190	56	9	58
	50 ft.	4	5	21	76	51	30	18	47	83	214	60	16	53

A further analysis of the results at Lake Cochituate shows the vertical distribution of the different classes of organisms to be as follows:

TABLE 52

NUMBER AND CLASSES OF ORGANISMS AT THE SURFACE AND BOTTOM OF LAKE COCHITUATE. 1895

Standard Units per cc.

	Diatomaceæ	Chlorophyceæ	Cyanophyceæ	Protozoa	Rotifera	Miscellaneous	Total
Surface.....	144	79	108	17	3	4	355
Bottom, 60 ft.	160*	16	67	10	1	99†	353

* If the dead and empty cells were excluded this figure would be much lower.

† Chiefly Crenothrix.

Birge and Juday have investigated the vertical distribution of plankton in a number of American lakes. Figures 71, 72 and 73 showing the vertical distribution of organisms in Hemlock, Conesus and Cayuga Lakes are taken from their "Limnological Study of the Finger Lakes of New York." The curves for phytoplankton indicate the usual dis-

tribution of these chlorophyll-bearing organisms. They are confined chiefly to the upper strata of water (the epilimnion or zone of circula-

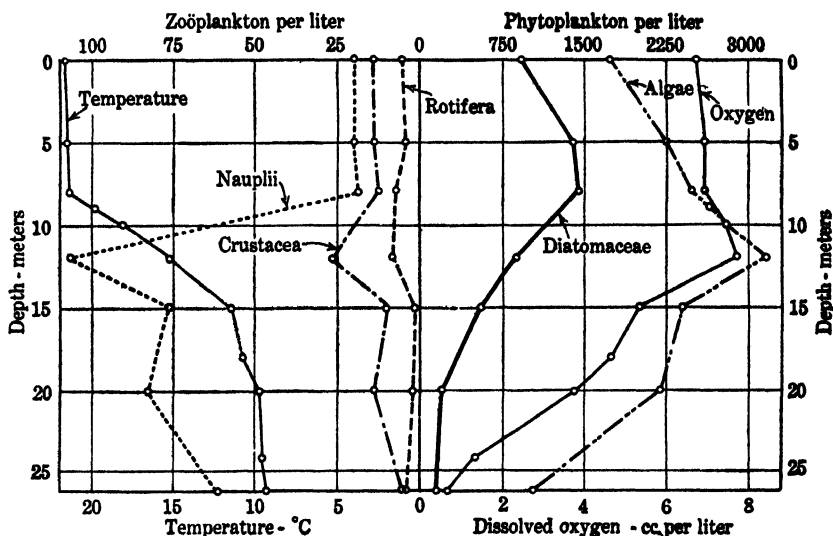


FIG. 71. — Vertical Distribution of Plankton Organisms in Hemlock Lake, Aug. 23, 1910. Predominant Forms: Diaptomus, Ceratium, Cœlosphærium, and Asterionella. After Birge and Juday.

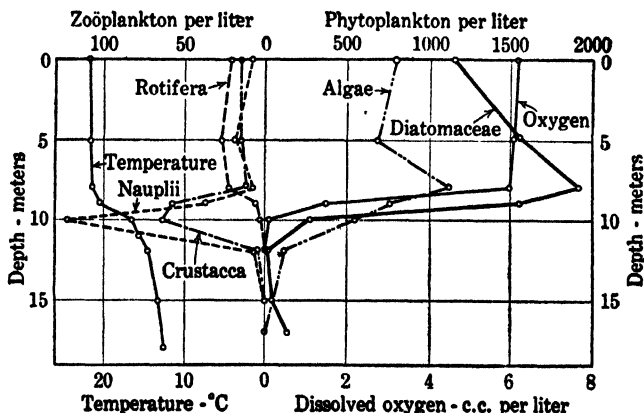


FIG. 72. — Vertical Distribution of Plankton Organisms in Conesus Lake, Aug. 25, 1910. Predominant forms: Cyclops, Polyarthra, Ceratium, Cœlosphærium, and Fragilaria. After Birge and Juday.

tion) where light conditions are favorable for their photosynthetic activities. The accumulation of synthetic organisms at the thermocline (transition zone) is reflected in the higher concentration of dis-

solved oxygen. Some of the algae, such as *Oscillatoria*, seem to adapt themselves to a saprophytic existence and then maintain themselves in deeper water. In general, however, the occurrence of large numbers of phytoplankton in the hypolimnion (zone of stagnation) should be

regarded as an indication of their senility.

The curves for zooplankton show two types of distribution. One as exemplified by the catches in Hemlock Lake (Fig. 71) in which all strata are well populated; the other as obtained in Conesus and Cayuga Lakes (Figs. 72 and 73) in which the lower waters are but sparsely inhabited either by reason of lack of oxygen (Conesus Lake) or due to scarcity of food (Cayuga Lake).

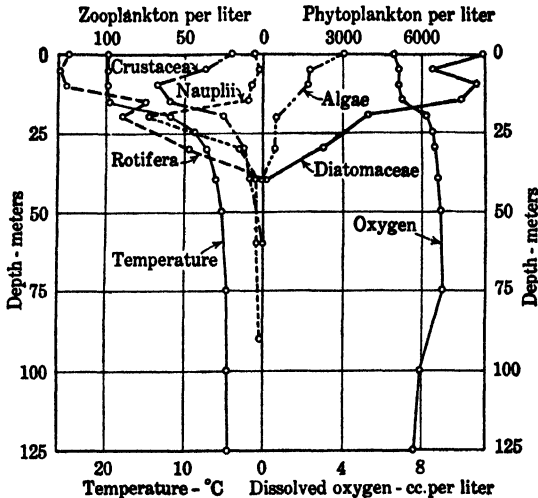


FIG. 73. — Vertical Distribution of Plankton Organisms in Cayuga Lake, Aug. 12, 1910. Predominant Forms: *Bosmina*, *Polyarthra*, *Ceratium*, and *Asterionella*. After Birge and Juday.

FREQUENCY OF OCCURRENCE OF DIFFERENT MICROSCOPIC ORGANISMS

The microscopic organisms that are found most commonly in the water supplies of Massachusetts taken from lakes or storage reservoirs are given in the following table, arranged according to the usual system of classification and divided into groups according to their abundance and frequency of occurrence. The first group includes those genera which, in their season, are often found in large numbers; the second group includes those which are found but occasionally in large numbers; the third, those which often occur in small numbers; the fourth, those which are rarely observed. This division, while not wholly satisfactory, enables one to separate the important from the unimportant forms. As observations multiply, the list may be extended and some genera may be changed from one group to another. The organisms printed in heavy type have given trouble in water supplies, either by producing odors or by making the water turbid and unsuitable for laundry purposes.

TABLE 53

FREQUENCY OF OCCURRENCE OF DIFFERENT MICROSCOPIC ORGANISMS

Massachusetts Experience

A. — Plant Organisms

Frequency of Occurrence	Cyanophyceae	Chlorophyceae	Diatomaceae	Schizomycetes and Other Fungi
Commonly found in large numbers	Anabaena Clathrocystis Coselosphaerium Microcystis	Chlorococcus Protococcus Scenedesmus	Asterionella Cyclotella Melosira Synedra Tabellaria	Crenothrix
Occasionally found in large numbers	Aphanizomenon Chroococcus Oscillatoria	Ankistrodesmus Coelastrum Cosmarium Palmella Pandorina Polyedrium Staurastrum Volvox	Diatoma Fragilaria Nitzschia Stephanodiscus	Didymohelix Sphaerotilus dichotomus
Commonly found in small numbers	Aphanocapsa	Closterium Desmidioidium Euastrum Eudorina Gonium Micrasterias Ophiocytium Pediastrum Sphaerososma Staurigenia Tetraspora Tribonema Ulothrix Xanthidium	Epithemia Gomphonema Navicula Stauroneis	Beggiatoa Leptothrix Molds
Occasionally observed	Glaucocapsa Lyngbya Merismopedia Microcoleus Nostoc Rivularia Sirostephon Tetrapedia	Arthrodesmus Bambusina Botryococcus Characium Chaetophora Cladophora Dactylococcus Dictyosphaerium Dimorphococcus Draparnaldia Glaucocystis Hyalotheca Mesocarpus Nephrocystium Penium Selenastrum Sorastrum Spirogyra Stigeoclonium Tetmemorus Zygnema	Achnanthes Amphiprora Amphora Bacillaria Cocconeis Cocconeis Cocconeis Cymbella Diadema Enconema Eunotia Grammatophora Himantidium Isthmia Meridion Odontidium Orthosira Pinnularia Pleurosigma Schisonema Striatella Surirella Tetracyclis	Achlya Leptomitosis Saprolegnia

TABLE 53. — *Continued**B. — Animal Organisms and Miscellany*

Frequency of Occurrence	Protozoa	Rotifera	Crustacea	Miscellaneous
Commonly found in large numbers	Cryptomonas Dinobryon Peridinium Synura Uroglenopsis			
Occasionally found in large numbers	Bursaria Chloromonas Glenodinium Mallomonas Raphidomonas			
Commonly found in small numbers	Actinophrys Amoeba Anthophysa Ceratum Cercomonas Codonella Epistylis Monas Tintinnus Trachelomonas Vorticella	Anuraea Conochilus Polyarthra Rotifera Synchaeta	Bosmina Cyclops Daphnia	
Occasionally observed	Acineteta Arcella Chlamydomonas Coleps Colpidium Cyphodera Diffugia Euchelys Euglena Euglypha Euplotes Glaucocoma Halteria Heteronema Naasula Paramecium Phacus Pleuronema Raphidodendron Stentor Synecrypta Trichodina Uvella Zoothamnium	Asplanchna Colurus Fosphora Floscularia Laciniularia Mastigocerca Microcodon Monocera Monostyla Noteus Sacculus Triarthra	Alona Cypris Diaptomus Sida	Acarina Anguillula Batrachospermum Chaetonotus Gordius Hydra Macrobrotus Meyenia Nais Spongilla

TABLE 54
 FREQUENCY OF OCCURRENCE OF DIFFERENT GROUPS OF MICROSCOPIC
 ORGANISMS
 Massachusetts Experience

Classification	Number of Genera				
	Commonly found in large numbers	Occasion-ally found in large numbers	Commonly found in small numbers	Occasion-ally observed	Total
Cyanophyceæ.....	4	3	1	8	16
Chlorophyceæ.....	3	8	14	21	46
Diatomaceæ.....	5	4	4	22	35
Schizomycetes and other Fungi.....	1	2	3	5	11
Protozoa.....	5	5	11	24	45
Rotifera.....	0	0	5	12	17
Crustacea.....	0	0	3	4	7
Miscellaneous.....	0	0	0	10	10
Total.	18	22	41	106	187

It will be observed that 187 genera have been recorded — 108 plants and 79 animals. Of these only 18 are commonly found in large numbers — 13 plants and 5 animals. 22 more are occasionally found in large numbers — 17 plants and 5 animals. 41 genera are frequently seen in small numbers, while 106 genera, or more than one-half of all are seen occasionally, some of them rarely. The most important classes are the cyanophyceæ, chlorophyceæ, diatomaceæ, and protozoa, as shown by the large number of genera and by their greater abundance. Furthermore, these classes include all the most troublesome genera that have been found in large numbers. There are 10 genera that are particularly troublesome because of their wide distribution, the frequency of their occurrence, and their unpleasant effects. They are *Asterionella*, *Anabæna*, *Clathrocystis*, *Cœlosphærium*, *Aphanizomenon*, *Dinobryon*, *Peridinium*, *Synura*, *Uroglenopsis*, and *Glenodinium*. This list seems like a short one when one considers the great extent to which microscopic organisms have caused trouble in water supplies.

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CHAPTER X

STORAGE OF WATER

No matter what the source of a water supply the water collected is commonly stored wholly or in part for varying periods of time before it is consumed. Storage permits more economic operation of water supply systems and insures an adequate supply of water at all times. In impounded supplies the water collected during periods of high run-off is stored to supply water during drier seasons. Water taken from rivers is stored in holding reservoirs or pumped into distributing reservoirs which form an integral part of nearly all distributing systems. Lakes and ponds are natural storage basins. Ground water systems have their collecting or distributing reservoirs. Water purification works are provided with pure water reservoirs. Rain water must be held in cisterns in order that water may be available when needed. It is during the period of storage that algæ troubles are observed most frequently hence this chapter deals with the effect of the conditions under which water is stored upon the growth of microorganisms. Most of the scientific principles underlying these effects have been discussed in the three preceding chapters on limnology. The present chapter deals in part with the operation of these principles in the practical problem of water storage, in part with certain limnological questions not previously discussed.

Sanitary Benefits of Storage. — Before the days of water chemistry and biology, when little was known of the purification processes that are ever active in bodies of water, when water was judged by its color, odor, taste, the presence or absence of “green scum,” and similar sensory evidences of apparent water quality, the expression “stagnant water” was often sufficient to create antagonism against a stored supply in the minds of the populace. “Running water purifies itself” was a slogan that seemed to exclude *per se* the possibility of standing water doing so too, and perhaps to a better degree. The reason for this perverse concept of sanitation is easily understood when we remember that standing water presents a favorable breeding ground for algæ and that microscopic growths, together with other influences operative in storage reservoirs, often produce conditions offensive to the senses of sight, smell, or taste.

With the genesis of modern sanitary science it was soon learned that

the evidence of the senses does not commonly form a reliable basis for judging the sanitary condition of water. The beneficial effects of storage were early recognized and storage was knowingly resorted to more and more to protect the health of water consumers as well as to meet the demands of rapidly growing cities for large volumes of water which could not be obtained economically in any other way. It was realized that the growth of microscopic organisms and other vegetation in reservoirs probably presented the only serious drawback to the use of water from lakes and impounded reservoirs; the interest of sanitary engineers in the plankton life of water dates back to this time.

The beneficial effects of storage upon the hygienic quality of water are due to several factors that may be summarized briefly as follows. Disease-producing organisms that may find their way into water do not encounter in it an environment favorable to their survival and far less to reproduction. Deprived of the requisite food substances they die according to the law of organic death, the number dying in each unit of time being proportional to the number surviving or nearly so. The longer the time elapsing between their passage from host to host, the fewer survive. The element of time introduced by storage, therefore, results in the gradual reduction of pathogenic bacteria. They are further decreased by sedimentation which is more active in quiescent water than running water. The presence of other microorganisms deprives the bacteria of food and many are ingested by predatory protozoa. Sunlight exerts its germicidal action at the surface of reservoirs and accounts for further destruction of pathogens. Taken together the effect of these various factors explains why long storage is a safeguard to the health of water consumers.

STORAGE OF SURFACE WATER

To obtain without purification a permanently safe and satisfactory surface water supply the rainfall must be collected quickly from a clean catchment area and stored in a clean reservoir.

This statement covers in few words all the factors that affect the quality of surface water. Just what constitutes cleanliness of catchment area and storage, however, is a question that involves the study of a great many agencies that in one way or another bring about changes in the composition of water. These agencies will, therefore, be discussed particularly in their relation to the growth of microscopic organisms.

Effect of Physiography of Catchment Area. — A clean catchment area may be defined as one upon which there is no source of pollution

and no accumulation of organic matter. The subject of pollution is of paramount importance, but will not be emphasized here, as its discussion leads more into bacteriology than into microscopy.

Indirectly, pollution affects the growth of microscopic organisms by contributing to the water food substances both living and dead upon which the plankton grow. The chief sources of pollution are agricultural lands, human habitations, and industries. The pollution of a catchment area, therefore, varies directly with the acreage of land under cultivation, and with the density of the population and its industrial activities. No catchment area is wholly free from organic matter which must eventually decompose. Even on clean gathering grounds grass dies, leaves fall and a thin layer of decaying matter is spread over the surface. This is repeated year by year. Normally the organic matter disappears by rapid oxidation, and if the ground slopes appreciably the rain that falls upon it runs off rapidly and absorbs little organic matter. If, however, the decaying vegetation has accumulated in deep layers and the ground is more nearly level the rain saturates or covers the organic matter with water; decomposition then takes place under different conditions and the water may become highly charged with organic substances and decay. This is the case in swamps.

Swamp Land. — The decaying organic matter in swamps furnishes food material for microscopic organisms, which in turn may render the water very disagreeable. Swamps, too, are breeding-places for many of the organisms that cause trouble in water supplies, and numerous instances can be cited where organisms that have developed in a swamp have been washed down into a storage reservoir, where they have rendered the water almost unfit for use.

Cedar Swamp, at the head of the Sudbury River of the Boston water supply, will serve as an example. During August, 1892, *Anabæna* developed abundantly in a small pond situated in the middle of this swamp. At one time the swamp water contained 8400 filaments (about 50,000 standard units) per cc. A heavy rain washed the chains of *Anabæna* down-stream, and on the fifteenth of August 2064 filaments per cc. appeared at the upper end of Sudbury Reservoir No. 2, a long and narrow body of water. On August 17 the water entering the basin contained but 600 filaments, and a week later *Anabæna* had disappeared from the inflow. In the mean time the growth of *Anabæna* continued down-stream like a wave, which passed successively through the basin, aqueduct, Chestnut Hill reservoir, and into the service-pipes. On August 22 *Anabæna* was first observed at the gate-house at the lower end of Reservoir No. 2; 647 filaments per cc. were counted. On the following day the organisms appeared at the terminal

chamber of the conduit at Chestnut Hill reservoir, where the presence of 326 filaments per cc. was recorded. In another week *Anabæna* became disseminated through this reservoir and was found in the service-pipes. Fortunately, the water from Reservoir No. 2 in passing to the city was mixed with water from other sources, so that by the time it reached the consumers *Anabæna* was not sufficiently abundant to cause complaint. After the first wave of *Anabæna* had passed through Reservoir No. 2 organisms of this genus began to increase throughout the basin, and the growth continued for several weeks. It became evident that the water from the swamp carried with it not only the *Anabæna* filaments themselves, but enough food-material to support their growth in the basin.

Organisms from swamps have frequently seeded storage reservoirs. Entering the reservoir in comparatively small numbers, the organisms often find in the quiet water conditions favorable to their development. Growths of some of the flagellates have thus been traced directly to seeding from swamps.

The effect of swamp areas upon the color of water has been referred to. Water from a clean gathering ground seldom has a color higher than 30 p.p.m. Higher colors can be generally traced to swampy land. The color of stagnant swamp waters is sometimes very high — often 300 and sometimes 500 or 700 p.p.m. From this it is readily seen that even a comparatively small percentage of swamp land upon a catchment area may have an important effect upon the color of the combined yield. If the color is much above 50 the water has an unsightly appearance, a distinct vegetable odor, and a sweetish and somewhat astringent taste.

A highly colored water means a water rich in organic matter. The effect of organic matter is to favor microscopic growths by providing food for development. The effect of color, however, is to reduce the occurrence of chlorophyllaceous organisms by absorbing light necessary for plant growth. Color, therefore, although indicative of favorable food conditions opposes photosynthesis. Large growths of algae, as a result, are more frequently associated with moderate colors than extremely high ones. (See Table 57.) Analytic organisms are not affected by color.

Ponds and Pools. — Small mill ponds and other imperfectly cleaned ponds or pools are also frequent breeding-places of microscopic organisms. Again the Boston water supply furnishes an example. A short distance above Sudbury reservoir No. 3 there were at one time several mill ponds. These were favorite habitats of *Synura* which was often found there in large numbers. When the water was let down-stream

through the mills or when heavy rains caused the ponds to overflow, *Synura* became numerous in the reservoir.

Summarizing the effect of the nature of the catchment area on water quality it is seen that in order to avoid the growth of troublesome organisms the water should be delivered from the collecting grounds *quickly*, and should not be allowed to stand in contact with organic matter in swamps or in shallow ponds or pools. As far as possible, therefore, the catchment area should be self-draining. The draining of swamps alone will make a vast improvement in the quality of the water delivered to the reservoir or intake.

Effect of Hydrography of Reservoir. — Many are the ways in which the natural characteristics of reservoirs can affect the growth of microorganisms and many are the attempts to measure and classify the relative values of these characteristics. In spite of the great amount of study that has been given to microscopic organisms, however, we are still far from formulating the laws governing their distribution. Why it is that a certain genus will grow vigorously in one pond and at the same time be absent from a neighboring one where conditions of existence are apparently as favorable, or why a form will suddenly appear in a pond where it has never before been seen we are still unable to say with certainty. Solution of such problems involves a far-reaching knowledge of the life history of the organisms, besides the effect of physical and chemical conditions upon their growth. The sciences of biophysics and biochemistry are yet in their infancy. Until they have been further developed many problems connected with microscopic organisms must remain unsolved.

Area. — The surface of a reservoir is the window through which sunlight penetrates into the plankton habitat. The greater the area therefore, the greater the opportunity for the growth of chlorophyll-bearing microorganisms.

Mere size in itself, however, has apparently little to do with the development of plankton. Algae are present in small as well as large bodies of water. To determine the effect of area the relative extent of the reservoir in proportion to the amount of water stored must be considered rather than the area itself. Other factors too come into play. The penetration of light into water varies with the color and turbidity of the water. The amount of radiant energy available for plankton growth therefore depends also upon the physical characteristics of the water. So many factors indeed are operative that it is almost impossible to gauge the influence of one factor such as area alone.

Depth. — It is well established that microscopic organisms are more abundant in the surface layers of lakes and reservoirs than in the deeper

strata. Reighard found 1.2 to 37.2 times as many organisms in the upper 5 feet of Lake St. Clair as in the remaining depths of 8 to 30 feet. Birge found 50 per cent, or more, of the crustacea in the upper 11 to 15 feet and over 90 per cent in the upper 33 feet of a 66 foot lake, and Ward reports 64 per cent of the plankton in the upper 7 feet. These facts, however, do not mean that the production of all types of plankton varies inversely as the depth of a reservoir. The fallacy of such a conclusion is shown by the following table.

TABLE 55
FREQUENCY OF ORGANISMS IN 41 SHALLOW AND 16 DEEP LAKES
Massachusetts Experience. 1900

Depth*	Number of Organisms per cc.	Per Cent of Lakes			
		Diatomaceæ	Chlorophyceæ	Cyanophyceæ	Protozoa
Deep	Often above 1000	50	19	12	12
Deep	Occasionally above 1000	12	12	6	0
Deep	Usually between 100 and 500	38	50	38	76
Deep	Always below 100	0	19	44	12
Shallow	Often above 1000	39	5	12	15
Shallow	Occasionally above 1000	14	22	22	17
Shallow	Usually between 100 and 500	32	51	29	56
Shallow	Always below 100	15	22	37	12

* Lakes of the Second Order are here called "deep lakes"; lakes of the Third Order "shallow lakes"; no lakes of the First Order are included.

There were 16 deep and 41 shallow lakes. Of the deep lakes 62 per cent at times had growth of diatoms above 1000 per cc. while of the shallow lakes 53 per cent had such growths. In no deep lakes were the diatoms lower than 100 per cc., while in 15 per cent of the shallow ones they were lower than this. It thus appears that heavy growths of diatoms are somewhat more likely to occur in deep than in shallow lakes. This is probably due to the influence of the spring and fall overturns which are more marked in deep bodies of water. The prevalence of grass-greens was similar to that of the diatoms though the difference was not so marked. 31 per cent of the deep lakes and 27 per cent of the shallow lakes at times had growths as high as 1000 per cc. The blue-greens and protozoa, on the other hand, inclined towards shallower water. In the case of the former, 18 per cent of the deep lakes and 34 per cent of the

shallow lakes at times had growths of 1000 per cc., while in the case of the latter the figures were 12 per cent and 32 per cent respectively. The importance of differentiating between the prevalence of different groups of organisms in measuring the effect of various factors is evident.

Shore-Line. — The shore forms a very important part of the aquatic environment. Here land and water come into most intimate contact; seepage and drainage waters enter; larger vegetation is most abundant; currents and other water movements are less swift; temperatures fluctuate more widely. Some of these conditions favor plankton growth, others oppose it. The relative effect will therefore vary in accordance with the dominant influences.

In order to compare the relative extent of the shore line of different bodies of water Seligo suggested the use of units called the "absolute" and "relative shore line developments." The absolute shore line development, according to Seligo's definition, is the length of shore line divided by the square root of the area, and the relative development is the absolute development in terms of the absolute development in a circle with unit radius. The effect of shore line development on plankton growths, however, is so masked by other factors that no generalization can be made.

Capacity. — The capacity of a reservoir is in itself of little moment so far as algæ growths are concerned. When it is taken into account as storage per square mile, number of days storage of the water, ratio of capacity to inflow or some similar unit, however, there seems to be a rather consistent inverse relation between the relative capacity and the growth of microorganisms. This does not mean that the capacity of the reservoir affects the organisms directly. Rather does it signify that the many factors that constitute the physical environment of lakes, ponds or reservoirs are reflected by the relative storage. This relationship is shown in Table 56. It is seen that the prevalence of microscopic organisms in the reservoirs of the Wanaque and Pequannock drainage areas decreases as the storage ratio increases.

Pockets. — When the bottom of a reservoir is uneven, water is apt to be left in small pools as the reservoir is drawn down. These pools are usually shallow and the water retained in them becomes warm and stagnant. It frequently supports rich cultures of organisms which are scattered through the reservoir when the pools overflow. As is true for the catchment area it is best to make the reservoir itself self-draining. Pools or pockets should therefore be provided with an outlet. If this is impossible it may be advisable to fill them up. The author once observed in a reservoir a pocket that was excavated to a considerable depth for the sake of removing all the organic matter at the bottom.

This pocket could not be drained, and during the summer it became the breeding-place of *Synura* and other organisms. It would probably have been better to have removed only a portion of the organic matter and to have covered the remainder with clean gravel or sand

TABLE 56

RELATION BETWEEN STORAGE RATIO AND PREVALENCE OF MICROSCOPIC ORGANISMS
Reservoirs of the Wanaque and Pequannock Drainage Areas in New Jersey
After Weston

Body of Water	Drain- age Area, Square Miles	Capacity, Million Gallons	Yearly Inflow, Million Gallons	Storage Ratio	Micro- Organ- isms, Standard Units per cc.
(1)	(2)	(3)	(4)	(3) ÷ (4) = (5)	(6)
Sterling Lake.....	5.1	3,586	2,360	1.52	93
Greenwood Lake.....	27.1	14,079	11,354	1.24	188
Canistear Reservoir....	5.6	2,407	2,625	0.92	366
Clinton Reservoir.....	10.5	3,518	4,935	0.71	558
Pequannock, total.....	63.7	11,808	29,890	0.39	518
Oak Ridge Reservoir...	27.3	3,982	12,800	0.31	569
Echo Lake.....	4.6	612	3,982	0.28	579
Forge Pond.....	14.7	7.5	7,090	0.011	652

Effect of Various Environmental Factors on the Occurrence of Anabaena.—

The statistics in Table 57 show the relation between the occurrence of a typical blue-green alga and some of the various factors previously mentioned.

Effect of Stagnation. — Water, as stated, should not be allowed to stand for any length of time in contact with organic matter and it is quite as bad for water to stand *over* a swamp as it is for it to stand *in* a swamp. The former indeed may be worse, for if the water is sufficiently deep decomposition of the organic matter at the bottom may take place in the absence of oxygen, when some of the resulting products are more easily taken up by the water. This brings us to a consideration of the so-called "stagnation effects." By stagnation is meant a continued state of quiescence of the lower layers of water in a lake or reservoir. It is caused by thermal stratification, as described in Chapter VII. During these periods of quiescence the water below the transition zone, i.e., the stagnant water, undergoes certain changes, the character and

amount of which vary with the nature of the water and especially with the presence or absence of organic matter at the bottom of the reservoir.

TABLE 57

EFFECT OF VARIOUS ENVIRONMENTAL FACTORS ON THE OCCURRENCE OF ANABÆNA

	Number of Lakes and Reservoirs	Per Cent of Lakes and Reservoirs Giving Stated Trouble from Anabæna		
		None	Slight	Much
Area of Lake, square miles:				
0-.1	48	50	29	21
.1-.25	29	31	21	48
.25-.50	16	19	37	44
.50-1.00	3	67	0	33
1.00-	5	20	40	40
Average depth, feet:				
0-10	24	71	25	4
10-20	34	29	21	50
20-30	4	25	0	75
Length of storage, days:				
0-50	7	71	15	14
50-100	3	33	0	67
100-200	7	29	57	14
200-500	21	29	19	52
500-	7	29	14	57
Color of water, p.p.m.:				
0-20	61	48	26	26
20-40	27	33	19	48
40-60	8	0	75	25
60-80	5	40	20	40
80-100	1	100	0	0
Population per sq. mile:				
0-10	29	52	17	31
10-50	31	49	26	25
50-100	20	35	40	25
100-200	11	36	28	36
200-	14	29	21	50

Physical and Chemical Effects. — Stagnation may be studied best in ponds where there is a considerable deposit of organic matter; of such ponds Lake Cochituate is an excellent example.

Lake Cochituate has a depth of 60 ft. near the gate house. At the bottom is found a layer of organic matter of unknown thickness which contains in its upper portion deposits of organisms and other organic

material transported by the water. The period of summer stagnation extends from April to November, and during these months deposits of organic matter accumulate at the bottom.

The changes that take place during the summer in the deep water strata of Lake Cochituate are shown in the following table (Table 58), in which analyses of the water at the surface and bottom are compared. The most conspicuous change is that of the color (see Fig. 74). While

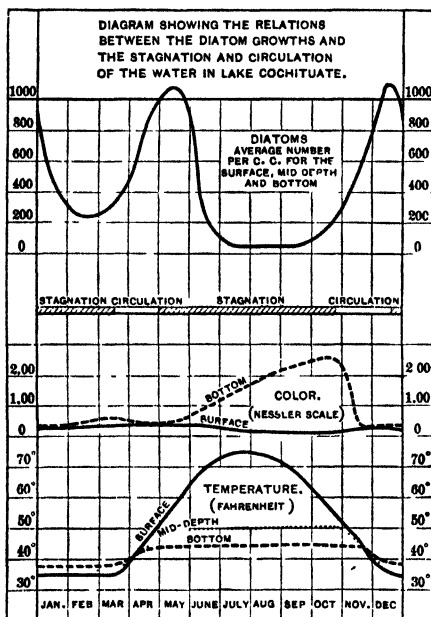


FIG. 74. — Stagnation Effects in Lake Cochituate.

under the influence of sunlight, the water at the surface is bleached, that at the bottom rapidly grows darker until, near the close of the stagnation period, it has a decided opalescent turbidity and a rich brown color that becomes even deeper when the water is drawn to the surface. The increased color is due to the presence of iron in the water. A considerable deposit of iron is found at the bottom. This is derived from the sedimentation of iron in combination with organic matter and from the precipitation of ferric hydrate as a result of oxidation in the upper layers. When, during the summer, the oxygen dissolved in the bottom water becomes exhausted, the ferric iron in the deposits gives up its oxy-

gen to the processes of decay and becomes reduced to the soluble ferrous state in which it is readily taken up by the water. When carried to the surface it becomes oxidized to the insoluble ferric state, deepening the color of the water for a time, but later precipitating as a brown floc and leaving the water with little color.

During stagnation important changes also take place in the organic matter in the lower layers. The amount of organic matter in the water increases by sedimentation from above and by solution from the ooze at the bottom. Albuminoid ammonia increases. Decomposition of organic matter takes place. Dissolved oxygen disappears and nitrates and iron become reduced. Free ammonia and nitrites increase, and the content of free carbonic acid becomes much greater. After the

TABLE 58
CHEMICAL ANALYSES* OF WATER AT THE SURFACE AND BOTTOM OF LAKE COCHITUCATE DURING THE PERIOD
OF SUMMER STAGNATION, 1891
Parts per Million

Date	Temperature, °F.		Color†		Albu- minoid Am- monia		Free Ammonia		Nitrites		Nitrates		Fixed Solids		Hard- ness		Iron		Manga- nese		Silica		Dis- solved Oxy- gen		
	Surface	Bottom	Surface	Bottom†	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom	
Apr. 3.....	48.6	43.2	.36	.36	.18200600130	18.6	During period of circulation
Apr. 8.....	25.0	27.56	.7	.4	.5	5.9	3.0	
June 6.....	69.1	44.4	.33	.88	.170	.190	.016	.224	.002	.002	.20	.21	30.5	36.5	17.4	18.4	.3	1.6	.5	.6	...	4.4	During period of stagnation
July 17.....	73.9	43.4	.21	1.51	.174	.212	.004	.430	.002	.004	.12	.08	24.5	40.0	17.6	19.5	.6	5.0	.3	.7	
Aug. 19.....	74.4	43.7	.19	2.56	.156	.262	.012	.600	.004	.006	.03	.02	21.5	51.5	18.9	21.5	1.1	9.7	.4	2.0	79	0	During period of circulation
Sept. 23.....	71.2	44.3	.13	2.93	.134	.244	.002	.736	.002	.005	.02	.02	35.5	58.0	18.2	19.8	1.1	9.6	.4	2.1	3.1	8.6	100	0	
Oct. 23.....	57.4	43.7	.16	3.7535286000300	...	64.5	...	22.0	During period of circulation
Nov. 2.....	45.4	44.6	.33	.34	.14404400120	...	35.0	...	19.0	
Dec. 2.....	39.3	39.9	.33	.37	.21203200312	...	32.0	...	18.0	

* Made by Dr. F. S. Hollis.

† Nessler Scale.

‡ After standing several hours.

supply of readily available oxygen (dissolved oxygen) has become exhausted, putrefaction through the agency of anaërobic bacteria takes place and the water acquires offensive odors. Increasing amounts of mineral matter are taken up from the bottom by the lower layers of water. This is true not only of iron, but also of silica, manganese, and some of the calcium and magnesium salts. At the same time, the bacteria below the transition zone multiply.

These stagnation effects are observed only below the transition zone. The relative changes that occur at different depths are well indicated by the amount of dissolved oxygen that is found, and the progress of the changes through the season may be studied by a series of such observations. Elaborate studies upon this subject have been made at Jamaica Pond by the Massachusetts State Board of Health for the details of which the reader is referred to the Special Report of 1890 on "Examination of Water Supplies," and to the Annual Reports for 1891 and 1892.

Biological Effects. — The effect of stagnation upon microscopic organisms has been referred to. In deep reservoirs relatively little microscopic life exists below the transition zone. The ooze at the bottom is largely an accumulation of dead organisms. The few living organisms that are found there are largely bacteria, fungi, protozoa and crustacea, organisms that are saprophytic and play the part of scavengers. During stagnation, however, the water at the bottom acquires a supply of food material — both organic and mineral — suitable for microscopic life. After stagnation ceases and circulation begins, this food material is carried to the upper regions where, in the presence of light and oxygen, the plankton is able to utilize it. Diatoms in particular depend upon the food supply acquired by the water during periods of stagnation (see Fig. 74).

The stagnation of a pond that has bottom deposits of organic matter affects the quality of the water both directly and indirectly during circulation; i.e., when the bad water at the bottom is carried to the surface. The direct effects are rising color, increasing organic matter, and frequently unpleasant odors. The indirect effects are odors caused by the growth of organisms that has been stimulated by the availability of food materials.

Stagnation in Reservoirs at Panama. — Mr. John R. Downes, Physiologist to the Isthmian Canal Commission, has described the stagnation of the reservoirs of the water supplies at Panama. Here stagnation occurs even though the reservoirs are relatively shallow and the temperature of the water is high. For example, in the Cocoli reservoir, 9 feet deep, the temperature on one occasion was 83° F. at the surface and

80° F. at the bottom, yet the dissolved oxygen varied as follows: Surface, 8.6; 5 feet, 6.2; 7 feet, 0.8; 9 feet, 0.0 parts per million. In Carabali reservoir, 12 feet deep, there was no dissolved oxygen below 8 feet. In Comache reservoir, 26 feet deep, there was none below 14 feet. At certain seasons of the year there are periods of overturn as elsewhere. This stagnation of the bottom water has been the cause of some bad tastes and odors in the water supplies. Algæ occur in these waters but apparently do not cause as much trouble as one might suppose.

Effect of Organic Matter Found on Reservoir Bottoms. — The disagreeable effects of stagnation that have just been discussed are influenced by the depth of the reservoir only in so far as depth affects thermal stratification and with it the length of the stagnation period and the volume of water that becomes stagnant. Of greater importance in producing offensive stagnation effects are the quality of the water stored and the amount and character of the organic matter at the bottom. If the water itself is relatively free from organic substances and if the bottom of the reservoir contains no organic deposits, stagnation effects will be reduced. Thus it has been found that in the Wachusett reservoir of the Boston water supply, where all organic matter was carefully removed from the bottom before the reservoir was filled, the dissolved oxygen at the bottom does not become exhausted during stagnation periods although it is appreciably reduced in amount. The author once collected a sample of water from Lake Champlain at a depth of nearly 400 ft. The temperature was 39.2° F. — i.e., maximum density — and the water was probably in a state of permanent stagnation. The sample nevertheless was bright, clear, colorless and without odor. The material on the bottom was found to be almost perfectly clean gravel.

In new reservoirs much of the organic matter found on the reservoir bed is derived from the vegetation and swamp muck that becomes inundated when the reservoir is filled. In old reservoirs most of the bottom deposits are due to sedimentation from the supernatant water. Examination of the topsoil in the Ashokan reservoir of the New York water supply before it was filled showed it to contain from 5 to 10 per cent of organic matter over the major portion of the reservoir bottom; only limited areas tested over 25 per cent of volatile matter. The depth of muck was less than 5 feet in the greater part of the area but did reach 20 feet in small sections.

The organic matter originally present on the reservoir bottom naturally undergoes decay as soon as the reservoir is flooded and is then gradually consumed. As a result it exerts in the course of time a decreasing effect upon the supernatant water. This is illustrated in

Fig. 97, Chapter XIII, in which the color and plankton growths of Massachusetts reservoirs are plotted in relation to the years elapsed since the reservoir was filled. In the course of time the organic matter would disappear entirely from the reservoir bed were it not for the fact that fresh deposits are formed by sedimentation from the water itself.

The extent to which bottom deposits accumulate is shown in Table 59. The figures in this table were obtained from an examination of several artificial distribution reservoirs in New York City. The reservoirs were either originally clean or possess a concrete bottom.

TABLE 59
BOTTOM DEPOSITS IN NEW YORK DISTRIBUTING RESERVOIRS, 1906
After Hazen and Fuller

Name of Reservoir	Depth below Surface of Water, Feet	Loss on Ignition, Per Cent	Microscopic Examination
Central Park	19.80-19.99	16.08	Vegetable débris, numerous algæ, diatoms.
Central Park	37.65-38.14	16.19	Numerous diatoms, Nitzschia, Synedra, etc. Large numbers of algæ (probably Ulothrix).
Central Park	38.92-39.54	18.08	Many algæ, diatoms, amorphous vegetable matter.
Williamsbridge	38.45-38.72	18.44	Diatoms, amorphous vegetable matter, algæ filaments in large numbers.
Jerome Park	22.6 -23.8	22.98	Residue consists almost entirely of algæ and diatoms, very little organic débris.

Since there was no organic matter in the reservoirs when they were filled it follows that the bottom deposits were derived from the suspended matter in the water stored in them and from organisms growing in the water. In the case of Jerome Park the deposits had accumulated in 13 months. Once deposits such as these are formed, it stands to reason that the organic matter originally present on the reservoir bed is of little influence upon the quality of the water. Comparisons of these deposits with the topsoil of Ashokan reservoir show that they contain relatively more organic matter as measured by the *loss on ignition* test.

In shallow reservoirs organic matter will also cause a deterioration of the water stored, even if there is no summer stagnation. The water

at the bottom becomes warm and decomposition goes on rapidly. The products of decay taken up by the water support the growth of organisms — particularly of blue-green algæ. Moreover, during the winter when the surface is frozen these shallow ponds grow stagnant and the conditions become similar to those in deep ponds. After periods of winter stagnation, shallow ponds often contain heavy growths of diatoms. Organic matter at the bottom of shallow reservoirs and on the shores of deep ones affects the quality of the water also by supporting larger aquatic vegetation.

Effect of Vegetation. — In studying the effect of vegetation upon the quality of water in lakes and reservoirs, we must distinguish between five classes of vegetation.

1. Plants without attachment, as *Lemna* (duck-weed) and *Ceratophyllum* (hornewort).
2. Plants attached to bottom and wholly submerged as *Vallisneria* (Eelgrass) and *Flodea* (Water weed).
3. Plants attached to bottom but only partially submerged as *Nymphaea* (Water lily) and *Bidens beckii* (Water marigold).
4. Swamp plants rooted in the substratum with emergent leaves but able to endure inundation and temporary submersion.
5. Land plants.

The unattached water plants obtain their food wholly from the water, while those that are rooted obtain much of their nutriment (dissolved salts and gases) from the soil. The submerged fixed plants draw upon the water for additional sustenance, and emergent vegetation derives some of its food material (carbon dioxide) from the air. Inundated swamp and land plants make no use of the food material contained in the water proper.

It is evident that those plants which derive their food materials, wholly or in part, from the water will deprive the microscopic organisms of much of their nutriment and thus prevent the development of phytoplankton. In general, therefore, other things being equal, lakes and reservoirs free from larger aquatic vegetation produce more plankton than those rich in such vegetation. This is shown in the table on the following page.

The ratio of plankton production in vegetation-rich to that in vegetation-poor water shows a definite excess of plankton in the latter type of water. The ratio becomes especially large from August to November, the months of greatest vegetation in water.

Swamp and land plants have no direct influence upon microscopic growths and the effect of attached emergent water plants is probably only slight.

TABLE 60

COMPARISON OF PLANKTON GROWTHS IN VEGETATION-POOR AND VEGETATION-RICH WATERS

After Kofoid

Cubic centimeters of plankton per cubic meter of water

	Vegetation-Rich		Vegetation-Poor		Average Ratio
	Quiver Lake	Dogfish Lake	Thompson's Lake	Phelps Lake	
Jan.....	.27	.53	3.79	3.29	1 : 9
Feb.....	.67	1.10	1.27	5.68	1 : 4
March....	.77	1.96	2.96	5.68	1 : 3
April.....	7.26	10.50	14.40	11.77	1 : 1.5
May.....	6.85	5.79	29.59	25.33	1 : 4
June.....	1.28	1.75	10.66	11.40	1 : 7
July.....	.78	1.95	4.74	8.50	1 : 5
Aug.....	.77	2.51	6.19	58.12	1 : 20
Sept.....	.77	2.39	5.37	47.25	1 : 17
Oct.....	.69	3.05	10.64	27.68	1 : 10
Nov.....	.23	2.64	6.39	41.57	1 : 17
Dec.....	.63	3.76	3.68	21.96	1 : 6
Weighted Monthly Average	1.75	3.16	8.26	22.35	1 : 6

In shallow reservoirs water weeds sometimes propagate so rapidly that they themselves constitute a serious nuisance. Most of the aquatic plants are perennial; few depend upon seed reproduction, although many of the flowering plants fruit and produce seeds. The usual method of propagation is through vegetative reproduction by means of runners, tubers, buds, or stem fragments. Vegetative reproduction is relatively slow, and seed reproduction is successful only when the seeds are deposited on favorable soil. Ordinarily, therefore, the growth of water weeds is limited. Under favorable conditions, however, propagation may become very rapid. *Vallisneria spiralis* according to Ritchie multiplied at an extremely high rate in a shallow Australian reservoir that was drained at seeding time. Once established, the eelgrass was very difficult to remove. *Elodea canadensis* too has given trouble in many lakes and reservoirs. At times it develops in such masses that it becomes impossible to row a boat through the aquatic meadow that

it creates. No wonder it was called the "American waterpest" when it first established itself in certain German lakes. *Lemna*, *Potamogeton*, *Chara* and other larger aquatic plants, together with attached algæ, will also flourish so prolifically at times as to change the shallower reaches of lakes and reservoirs into veritable fresh-water "Sargassos".

The death and decay of vegetation in lakes and reservoirs injures the quality of the water by increasing the organic matter and producing food material for microorganisms. While swamp plants are able to endure inundation, land plants die quickly when submerged. In reservoirs that are drawn down for prolonged periods land plants tend to grow on the exposed reservoir bottoms wherever these are covered with organic matter and are able to support vegetation. When the reservoirs are filled again the plants are inundated, die and decay. By wind and wave action the organic débris is sometimes carried out from shore and furnishes food to hosts of animalcules while yielding valuable mineral salts which are utilized by the phytoplankton.

The larger aquatic vegetation often serves as a support for the more lowly organized forms, usually algæ and other sessile organisms that would be unable to live as free-swimming individuals. This is illustrated in Figs. 75 to 77 in which algæ are shown attached to the stems and leaves of larger aquatic plants. The aquatic plants also afford shelter for the protozoa and other zoöplankton that live upon the organic matter that accumulates in the protected waters of the aquatic meadows. The oxygen produced by the plants keeps the water well aerated, and favors the development of animal forms that would otherwise perish in the waters above the decaying organic matter.

Effects of Plankton on Bacteria. — There exist in all bodies of water rivalries both active and passive among the various groups of organisms. The struggle for existence becomes more intensified the lower the scale of life and manifests itself in relatively rapid changes in the micro-flora and fauna. The changes that occur in water are discussed more fully in the chapter on "Self-Purification of Streams" for the reason that the various zones of existence are more clearly defined in polluted streams, and cause and effect are more readily differentiated.

The succession of dominant species observed in water may be due to active warfare between the individuals as exemplified by bacteriverous protozoa and other zoöplankton, or it may be caused by changes brought about in the environment as a result of the growth or death of certain organisms. The effects of plankton growths upon bacteria are of especial interest to sanitary engineers; cases are on record where otherwise objectionable growths of microorganisms definitely improved

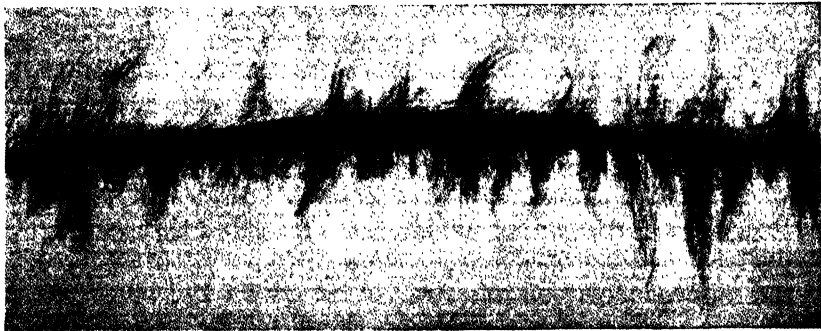


FIG. 75. — *Spirogyra* on Eelgrass.

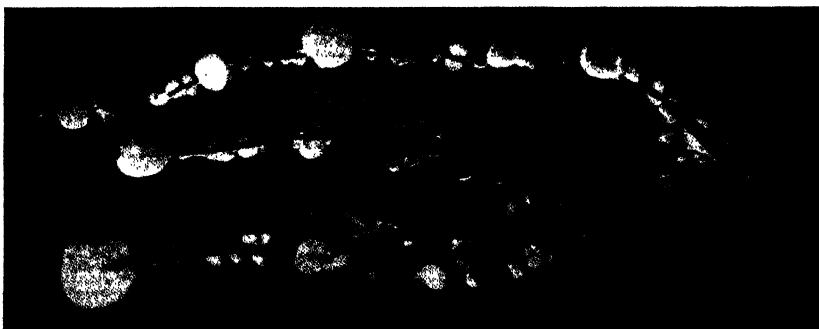


FIG. 76. — *Rivularia* on *Potamogeton*.



FIG. 77. — Minute Algae on *Potamogeton*.

the quality of drinking water. One of these is taken from the records of Mt. Prospect Laboratory.

Baiseley's Pond, one of the sources of water supply of Brooklyn, is fed by a number of streams which are more or less polluted. During August, 1899, a very large growth of *Clathrocystis* was recorded. At the same time bacteriological examinations of the inflowing streams showed

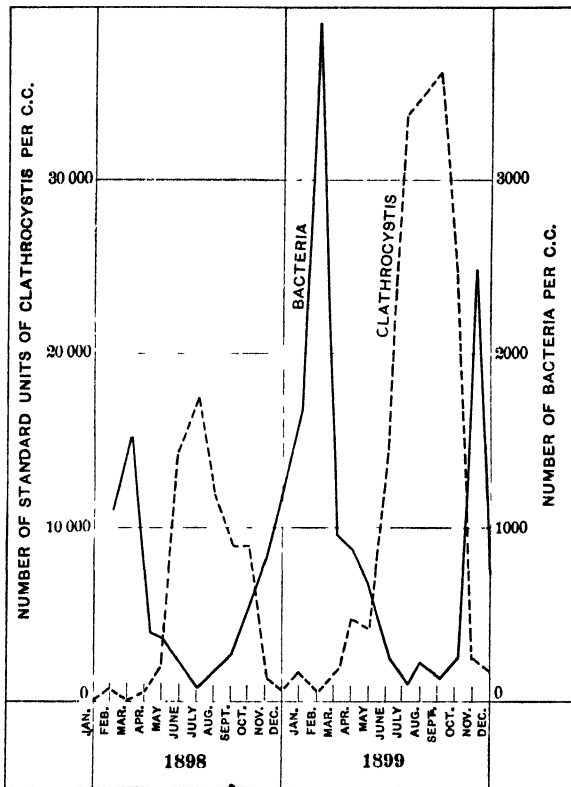


FIG. 78. — Effect of *Clathrocystis* upon the Numbers of Bacteria in Baiseley's Pond, Brooklyn Water Supply.

1000 to 1700 bacteria per cc. whereas less than 50 bacteria per cc. remained in the water at the lower end of the pond. A study of the records for the years 1898 to 1899 showed that the number of bacteria varied inversely with the number of *Clathrocystis*. (See Fig. 78). This inverse correlation was probably due to the fact that the growth of *Clathrocystis* used up the carbon dioxide of the water and rendered the environment unfavorable to the existence of bacteria. The effects upon bacteria of changes in the hydrogen ion concentration of their environ-

ment are well established as are the fluctuations in pH with variations in the carbon dioxide content of water. The lime process of disinfection is based upon these relationships.

Strohmeyer at Hamburg obtained laboratory evidence of the influence of algæ upon bacteria. Using cultures of *Enteromorpha*, a member of the green algæ, he obtained the following results.

TABLE 61
EFFECT OF *ENTEROMORPHA* ON BACTERIA

Date	Time	Number of Bacteria per cc.	
		Enteromorpha Present	Enteromorpha Absent
July 4	11.30 A.M.	145	108
" 4	2.00 P.M.	160	144
" 4	6.00 P.M.	152	243
" 5	8.30 A.M.	1100	5,900
" 5	2.00 P.M.	180	26,000
" 5	6.30 P.M.	7	50,000
" 6	9.00 A.M.	24	63,000
" 6	7.30 P.M.	0	80,000

While the phytoplankton affect bacteria indirectly by rendering their environment unfit, many of the zoöplankton destroy bacteria by preying upon them for sustenance. A large variety of protozoa, for example, obtain food by ingesting bacteria. Purdy and Butterfield have investigated this antagonism between protozoa and bacteria. The results of one of their experiments are shown in Fig. 79. The reduction in bacteria in the presence of protozoa is substantial, as is their increase in number following the dying-out of the protozoa. This rise in bacteria following the death of plankton, both animal and plant, is due to two factors (1) the removal of the plankton antagonism and (2) the increased food substances provided by the decaying organisms. At Cambridge the destruction of microorganisms in Fresh Pond Reservoir by means of copper sulphate always results in the immediate increase in bacteria from about twenty to several hundred per cc.

Effect of Wind and Waves. — Some microscopic organisms are extremely fragile and are readily broken up by agitation. Of these notably the filamentous algæ develop well only in fairly quiet water. As pointed out by Dr. Drown this explains why many forms of algæ are not

found in rivers. In the larger lakes and reservoirs wind and wave action are often sufficient to destroy or prevent microscopic growths. At Ludlow reservoir (Springfield, Mass.) large growths of *Anabaena* have

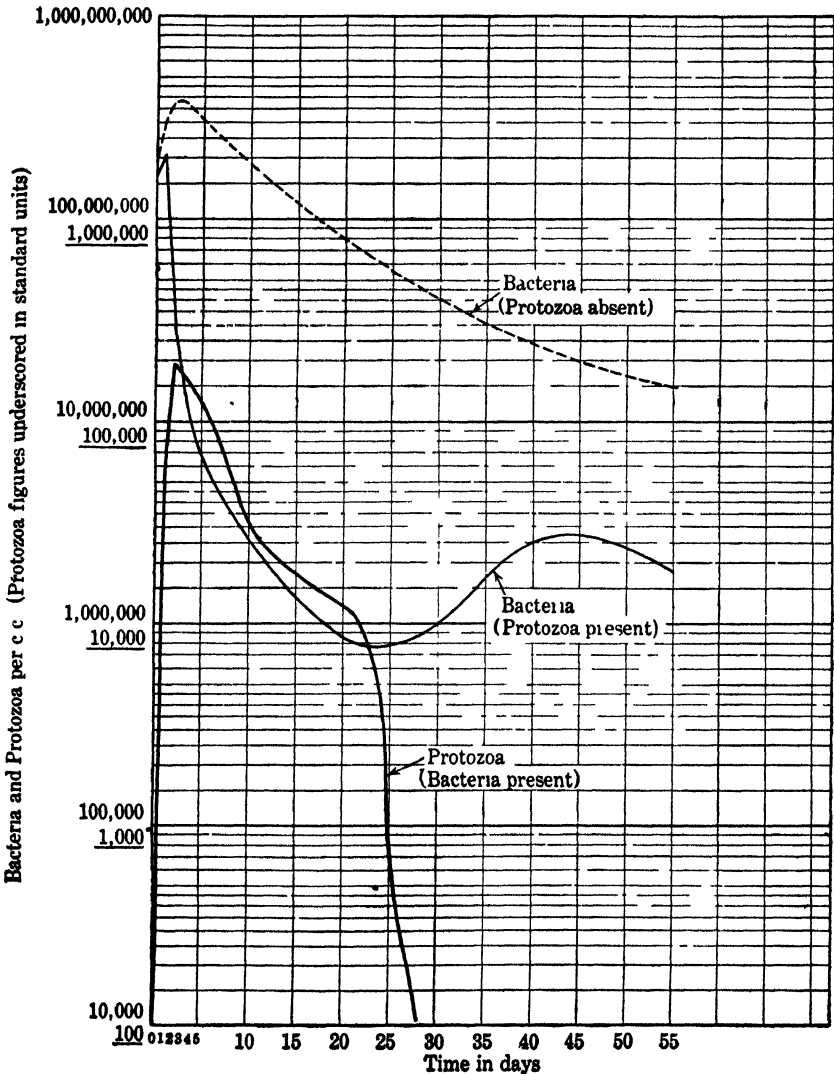


FIG. 79 — Growth of Bacteria in Sterile Tap Water in the Presence and Absence of Protozoa. After Purdy and Butterfield.

been eliminated by a single windy day. The filaments are broken up, the cells settle and die, and weeks often elapse before appreciable growths are again observed.

TABLE 62
MICROSCOPIC ORGANISMS IN THE RESERVOIRS OF THE CROTON WATER SUPPLY OF NEW YORK CITY
Compiled in 1907

Lake or Reservoir	Microscopic Organisms			Drainage Area, Square Miles	Area of Water Surface, Square Miles	Average Depth, Feet	Maximum Depth, Feet	Storage Capacity, Days Flow	Swamp Area on Water- shed, Square Miles	Swamp Area, Per Cent of Watershed	Ratio of Swamp Area to Water Surface	Average Color of Water, Parts per Million	Population per Square Mile on Watershed	Storage Capacity, Million Gallons	Date when Reservoir was Put in Use
	Fre- quency Rating	Average Number. per cc.													
		Entire Year	Summer Months												
Middle Branch Res.	83	1732	2048	20.51	0.77	25	50	195	1.17	6.0	1.5	22	31	4005.0	1870
Kirk Lake.	81	1230	1132	2.84	0.16	17	25	198	0.88	6.0	0.8	41	65	565.0	1897
Muscoot Res.	72	1069	1239	14.45	1.10	29	64	460	0.48	2.0	2.0	22	50	6692.0	1895
West Branch Res.	70	1084	1010	18.83	2.02	23.8	46	535	1.00	4.0	0.77	26	47	7167.0	1893
Titicus Res.	59	1798	1271	22.80	1.30	26.4	75	315	5.38	7.0	6.0	30	38	4900.0	1891
Sodom Res.	48	807	799	73.23	0.89	26	68	67	4.70	3.0	2.7	33	74	1842
Croton Lake, Old.	46	763	890	158.7	1.75	90	0.18	5.0	0.28	25	56	4145.0	1891
Bog Brook Res.	43	778	771	3.67	0.64	31	50	1130	15	126	575.0	1870
Lake Mahopac.	36	447	227	1.03	0.88	85	560	0.45	0.2	1.0	30	27	2720.0	1873
Boyd's Corner Res.	10	284	444	21.43	.44	29.8	50	127	11	390	165.0	1870
Lake Gleneida.	3	219	348	0.58	.28	2.8	100	243	61	380.0	1870
Lake Gilead.	0	56	94	0.65	.19	9.6	100	585

TABLE 63
MICROSCOPIC ORGANISMS IN THE RESERVOIRS OF THE METROPOLITAN WATER SUPPLY OF BOSTON, MASSACHUSETTS

Reservoir	Microscopic Organisms			Year when Built or Put in Use	Drainage Area, Square Miles	Area of Water Surface, Square Miles	Maximum Depth, Feet	Average Depth, Feet	Storage Capacity, Days Flow	Area of Swamp Land, Square Miles	Swamp Area, Per Cent of Drainage Area	Ratio of Swamp Area to Area of Reservoir	Per Cent of Water Surface on Drainage Area	Color of Influent, Parts per Million
	Frequency Rating Entire Year	Ave. No. of Organisms per cc. Entire Year	Ave. No. of Organisms per cc. Summer Months											
Lake Cochituate.....	50	783	663	1848	18.87	1.23	72	4.35	23.1	3.56	6	90
Framingham No. 3.....	26	475	689	1878	5.40	.396	25	.15	220	.15	2.8	.38	7	40
Hopkinton.....	25	453	682	1894	5.86	.29	5585	14.5	2.93	5	150
Sudbury.....	24	461	649	1897	22.28	2.02	67	1.85	8.3	0.92	0	50
Whitehall.....	21	406	412	4.35	.94	1813	3.0	0.14	22	60
Wachusett.....	20	541	386	1905	118.23	6.56	129	46.2	534	3.55	3.0	0.96	6	50
Ashland.....	7	214	321	1885	6.43	.26	4952	8.1	2.18	4	140
Framingham No. 2.....	5	190	429	1878	28.50	.21	18	2.17	7.6	10.3	1	95

The larger the reservoir the greater the effect of wind on wave action. As a result small bodies of water are much more subject to growths than are large ones. The absence of agitation also helps to explain why so much plankton is found in coves that are sheltered from wind action and among water weeds that prevent violent agitation of the water. Conditions of shelter, furthermore, account for the fact that lake water that is almost free from microscopic growths will frequently support a large plankton population when it is stored in a wind protected reservoir. At Syracuse, N. Y., Burlington, Vt., and Cleveland, O., for example, objectionable growths have occurred in distributing reservoirs supplied with lake water that itself contained but few microorganisms.

The effect of wind upon water circulation and the incident changes in the distribution of organisms and their food materials have been discussed in the chapters on "Limnology".

Seeding of Reservoirs. — The difficulty has been mentioned of explaining why certain lakes and reservoirs continue for years and sometimes indefinitely without "algæ troubles" while other bodies of water, in all ways similar, have their annual crop of microorganisms. One reason for this difference in behavior must be sought in the seeding of the water with the vegetative cells or the spores of the organisms. The mode of seeding is not well established. The spores are conceivably carried by wind or water, by migratory birds, fish or mammals or by accidental transportation on vessels and other conveyances. Sometimes the spores originate on the same catchment area, sometimes they are carried over the divide. Heavy seeding is undoubtedly due to the overflow of swamps or the transfer of partially dried spores from swampy areas in the vicinity.

The freedom from growths is especially remarkable in many ponds and lakes of the South in which conditions of plankton existence seem to be ideal. Some open distributing reservoirs of municipal water supply systems are also astonishingly free from growths although the water may have the characteristics commonly associated with the requirements for abundant algal growths. Such records have been established for filtered water from both slow and rapid sand filter plants. Among reservoirs that have given no trouble may be mentioned the distributing reservoirs at Paterson, New Jersey; Watertown, New York; and Cambridge, Massachusetts.

Prevalence of Algæ in Large Stored Water Supplies. — The prevalence of algæ in large stored water supplies is well illustrated by the conditions obtaining in some of the reservoirs of Boston and New York before the control of algæ was as well developed as it is today.

Algæ in the Croton Water Supply of New York City. — The water supply of New York City is taken in part from artificial reservoirs and natural lakes on the Croton River catchment area. In no case was the soil removed from the sites before the reservoirs were filled. Prior to control of plankton by copper sulphate and chlorine very heavy growths accordingly occurred in all of the reservoirs and this was also true of the distribution reservoirs in Central Park. The water as delivered in the city usually had a taste and odor caused by these growths. At times it was very noticeable and most unpleasant.

The occurrence of growths of organisms in a number of the Croton reservoirs before control methods were instituted, together with various data that bear upon the problem are shown in Table 62.

Algæ in the Metropolitan Water Supply of Boston. — Most of the reservoirs of the Metropolitan Water Supply of Boston have been less troubled with algæ than the reservoirs of the Croton supply used to be. This may be seen by comparing Table 63 with the preceding. The difference is due in part to the fact that many of the Massachusetts reservoirs were stripped.

STORAGE OF GROUND WATER AND FILTERED WATER

Ground Water. — *Ground water must be stored in the dark in order to prevent the growth of microscopic organisms.*

Water that has passed through the soil usually carries gases and mineral matter in solution, some of which, as carbon dioxide and nitrates, form important parts of plant food. The growth of synthetic organisms however does not proceed in the absence of light. As long as the water remains underground, therefore, chlorophyllaceous plankton does not develop.

Growth of Organisms in the Light. — When ground water is stored in an open reservoir it is liable to become seeded with microorganisms and to deteriorate. Diatoms are especially apt to develop; their mineral content is greater than that of most plants; also they require much silica. Growths are more likely to occur in old reservoirs in which a flora or fauna has become established. In new reservoirs, as a rule, the littoral organisms develop first, growing on the sides or even on the bottom of the reservoir. After deposits of organic matter have accumulated on the bottom, and the conditions have become more favorable the limnetic forms follow.

Of the diatoms that occur in ground water exposed to the light *Asterionella* is by far the most troublesome. Some diatoms merely increase the turbidity of the water by their presence, but *Asterionella*

also produces a marked odor. In surface waters *Asterionella* develops most vigorously after the stagnation periods; this is probably true also in ground waters. Ground water is commonly stored only for the purpose of meeting the fluctuations in consumption during short periods of time. Sometimes ground water reservoirs act merely as equalizing reservoirs in which a single pipe serves both as inlet and outlet. Circulation in such reservoirs is very poor and the water often becomes stagnant. This is also the case when the inlet and outlet of distributing reservoirs are improperly located. An open reservoir may give no trouble until a layer of organic matter has accumulated on the bottom and the water in some way becomes seeded with organisms such as *Asterionella*. Thereafter growths of organisms occur regularly. If open reservoirs are to be used for the storage of ground water they should be kept clean and adequate circulation should be insured.

Growth of Organisms in the Dark. — Darkness is not always sufficient to prevent ground water or filtered water from deteriorating. There are some organisms that can live without light, and indeed prefer darkness. Of such a nature are the fungi (using the word in its broad sense as including those vegetable forms destitute of chlorophyll) and some of the protozoa and larger animal organisms.

Crenothrix is the most important organism of this character that affects ground water supplies. It is a small filamentous plant, the cells of which are but little larger than the bacteria. Its filaments have a gelatinous sheath colored brown by a deposit of ferric oxide and grow in tufts, sometimes matted together into a felt-like layer. Other organisms similar to *Crenothrix* are *Leptothrix*, *Didymohelix*, *Sphærotilus dichotomus* and *Clonothrix*.

Crenothrix is liable to occur in ground water rich in iron and organic matter and therefore frequently infests water obtained from wells driven in swampy land. It is sometimes observed in imperfectly filtered water. *Crenothrix* may grow in almost any part of a ground water system — in driven wells, filter galleries, reservoirs, and distribution pipes and develops especially luxuriantly about wood work.

Mixed Surface and Ground Water. — When a water supply is derived from both ground and surface sources, the chances of obtaining plankton growths in reservoirs become great. Surface water usually contains some microscopic organisms and their growth is stimulated by the food material in the ground water. Surface water also contains organic matter which will be deposited and increase the effects of stagnation. Growths of *Asterionella* are apt to occur. The water supply of Brooklyn, N. Y., presents an interesting example.

Brooklyn derives part of its supply from a number of small storage

reservoirs along the southern shore of Long Island and from driven wells and infiltration galleries along the line of the aqueduct. The well water is drawn from depths varying between 25 and 200 ft. Surface and ground waters become mixed in the aqueduct and are stored in the three basins of Ridgewood reservoir. The quality of the water from the different sources varies greatly. Some waters contain an abundance of organic matter; some have high free ammonia, nitrites, and nitrates; some have considerable iron; and one or two have high chlorine and hardness due to admixture of a small amount of sea water. All have carbonic acid. The catchment area is sandy, and the waters are rich in silica.

In 1896 *Asterionella* developed in Ridgewood reservoir in great abundance and thereafter reappeared at intervals which coincided in a general way with the spring and fall distribution of this organism. The pulses of growth also corresponded to some extent with the use of increased proportions of ground water. Before control methods were adopted the numbers of *Asterionella* recorded were at times very high — 25,000 or 30,000 per cc. For many years prior to the occurrence of these larger growths Ridgewood reservoir caused no trouble and the water supply bore an enviable reputation. It was not until a considerable deposit of diatoms and other organic matter had accumulated on the bottom of the basins and until the amount of ground water had risen to about 40 per cent of the total supply that the conditions became favorable for such enormous growths of *Asterionella*. Fortunately for the consumers, a by-pass around the distributing reservoir permitted the water to be pumped from the aqueduct directly into the distribution system, whenever *Asterionella* became abundant enough in the reservoir to cause a bad odor. During recent years copper sulphate has been used to control the growths.

Filtered Water. — Water that has been filtered resembles ground water, and microscopic organisms may develop in it in sufficient numbers to cause trouble. For this reason provision is generally made for storing filtered water in covered basins. Often, however, economy requires the use of existing uncovered reservoirs. These may become infested with microscopic organisms, which, however, seldom cause as much trouble in filtered water as in ground water exposed under similar conditions. Alum-treated water is somewhat more subject to growths than other purified waters, because the use of sulphate of alumina liberates in the water a certain amount of dissolved free carbonic acid (6.8 p.p.m. of CO_2 for each grain of alum per gallon of water) which favors the growth of phytoplankton. Iron and lime which are also used in coagulation do not possess this disadvantage. As a result of

oxidation, the effluent of slow sand filters usually contains larger amounts of nitrogen in the form of nitrates and their increase may encourage algal growths. The controlling factor in the storage of purified waters, however, is usually the length of time that the water remains in the reservoir. If the period is short growths are usually insignificant, but if the water is kept in the reservoir for many days algæ are likely to develop to a troublesome extent.

As an illustration of the effect of storage on filtered water the following figures taken from analyses of the Hudson River water at Poughkeepsie, New York, before and after slow sand filtration are interesting:

TABLE 64
GROWTH OF MICROSCOPIC ORGANISMS IN FILTERED WATER
Poughkeepsie, New York, 1903

Date	Microscopic Organisms per cc.	
	Raw Water	Filtered Water after Storage
April 23.....	60	1455
May 11.....	70	135
June 8.....	95	65
June 20.....	65	130
July 8.....	205	655
July 23.....	230	2440
August 6.....	185	2265

The growth of organisms in filtered water stored in the open and in the dark is further discussed in Chapter XIII.

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CHAPTER XI

RHEOLOGY

Rheology, sometimes called potamology, is that composite science which deals with the properties and characteristics of streams. Like limnology it includes a number of subjects that have no direct bearing upon aquatic microscopy. These are not discussed in this chapter which is confined to a brief consideration of those environmental influences which are more or less peculiar to streams and most affect microscopic life in flowing water.

Whereas microscopic organisms find in lakes and other standing bodies of water relatively stable conditions of existence the plant and animal life of streams is subjected to continuous changes in the constitution of its habitat.

Life in a swift-flowing stream is far more complicated and hazardous than in the shelter of a quiet backwater.

If we consider the stream as a whole the complexion of the environment is found to vary along its entire length. Channel contours change from place to place; shallow flowage and backwater alternate with deep pools and swift water. Current velocities vary; at times they are sluggish, at times turbulent. Different geological formations follow one another in quick succession; clean hard bottoms give way to soft mud deposits and *vice versa*. Tributary waters flowing into the streams from ground or surface sources modify the quality of the water and with it the food supply of river life. Waste waters from human habitations and industrial plants bring about sudden and often catastrophic changes. In this unstable environment only a few organisms that have sufficient powers of locomotion to swim against the stream or to withstand its transporting power can select their habitat. Organisms that drift with the current must take things as they come; they are subjected to many different influences as the water passes down stream. Attached organisms thrive or succumb in accordance with the nature of the changes that take place in the stretch of river that they have made their home.

If we confine our attention to a particular cross-section of stream we find that life in it, too, is exposed to varied influences which from time to time bring about profound disturbances in the environment. Stream levels rise and fall; currents fluctuate in intensity; the quality of the water passing by changes both physically and chemically. Under these

unstable conditions existence is precarious and it is readily understandable why in general microscopic organisms develop more abundantly in lakes and reservoirs than they do in streams.

The factors affecting plankton life in streams may be classified as physical, chemical and biological, as they have been already for standing waters. Many of the factors are identical under both conditions of existence, most are similar, only a few are distinctly different. The methods of rheological investigation are therefore essentially the same as those developed for limnological work. Largely the same apparatus is used and examination and recording proceed along similar lines. Interpretation of the findings, nevertheless, requires thorough appreciation of the conditions of existence encountered in flowing waters.

PHYSICAL CONDITIONS

Of the physical factors affecting the rheoplankton or, as it has also been designated, the potamoplankton, heat and light play with but slight variations the same rôle as in standing water. Wind action on the other hand becomes of small importance while the effects of currents and changing hydrographic conditions loom large.

Temperature. — As in the case of lakes the seasonal changes in water temperature produce variations in the growth of rheoplankton that may be characterized as spring, summer, and winter phenomena. In his study of the Illinois River Kofoid recorded the following average catches during the years 1894 to 1899.

TABLE 65
SEASONAL PRODUCTION OF PLANKTON
Illinois River, 1894 to 1899

Month	Surface Temperature, °F.	Plankton, cc. per m. ³	Month	Surface Temperature, °F.	Plankton, cc. per m. ³
Jan. . . .	32.8	0.21	July.	81.0	4.23
Feb.	32.7	0.23	Aug.	81.5	3.88
Mar.	40.5	0.27	Sept.	74.2	2.56
April. . . .	60.5	4.59	Oct.	57.6	1.71
May.	68.3	6.08	Nov.	43.0	0.88
June.	77.8	7.22	Dec.	35.2	0.71
			Year*	57.1	2.71

* Average of Monthly Averages.

The seasonal variations shown in Table 65 are not only due to changes in temperature but are caused in part by fluctuating hydrographic conditions. These operate in the Illinois River to produce a summer

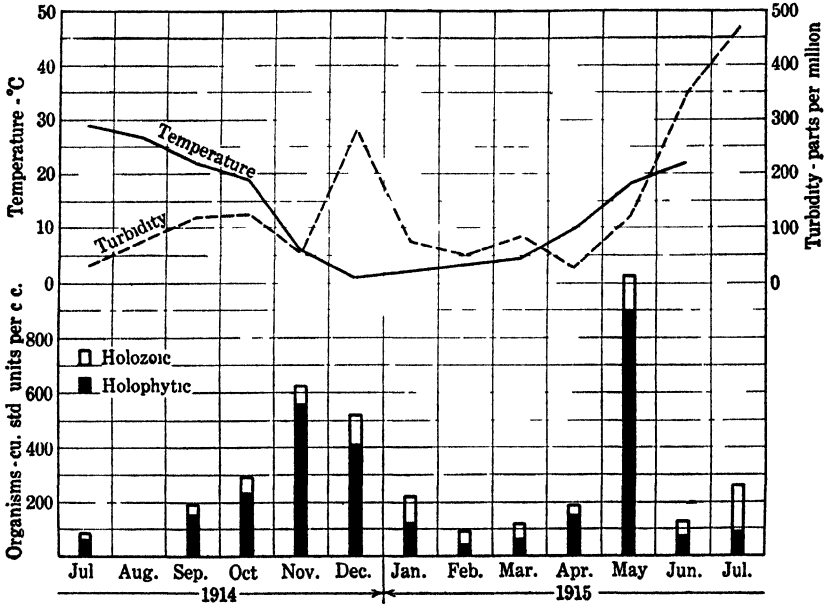


FIG. 80 — Seasonal Distribution of Plankton Organisms. Ohio River at Cincinnati. Monthly Averages 1914-1915. After Purdy; U. S. Public Health Bulletin 131.

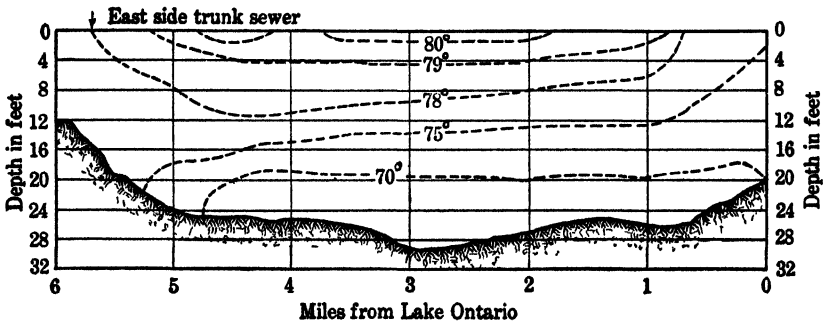


FIG. 81. — Thermal Stratification in °F of the Genesee River at Rochester, N. Y., July 15, 1912.

maximum in advance of the warmest weather, during the period of lowest flows. Somewhat similar conditions, observed by Purdy, for the Ohio River are shown in Fig. 80. The seasonal course of plankton growth is here modified considerably by the variation in the turbidity of the water.

Thermal stratification of streams is in evidence but the movement of the water usually prevents the vertical zonal differentiation observed in lakes. Figure 81 shows the summer temperatures at varying depths along the center line of the Genesee River as it approaches Lake Ontario. The isotherms in this stretch are affected both by the discharge of Rochester sewage into the river and by the backwater effects of the lake. True stagnation due to thermal stratification occurs probably only in the backwaters and deep pools of slowly moving rivers, but ice cover sometimes causes stagnant conditions even in streams, if they carry much organic matter.

Light. — The conditions of light in streams are varied from those existing in lakes largely by the presence of suspended matter which does not settle out as readily in flowing water as in standing water. In silt-bearing streams turbidity excludes much light, photosynthesis is curtailed and the growth of heliophilous organisms is discouraged. The amount and intensity of light received varies also with the orientation of the stream, its geographical location, and the season of the year.

Occlusion of light by turbidity is subject to fluctuations depending upon the amount of surface wash received from the catchment area and upon the current velocities which affect the silt-bearing power of the water. The seasonal variation in turbidity of the Ohio River at Cincinnati is included in Fig. 80 and exemplifies the effect of high turbidities on plankton catches.

Turbidity. — Turbidity is caused by organic and inorganic solids which are in a state of coarse and colloidal suspension; it is that property of water which renders indistinct the outline of objects viewed through it. The lack of distinctness is due to the refraction and absorption of light rays. Turbidity, therefore, is a factor of account in the ability of a stream to develop heliophilous organisms. In most lakes and ponds there is only slight interference from suspended particles to the passage of light.

Both submerged plants and rheoplankton are affected by turbidity. Low values do not prevent photosynthesis except at considerable depths, but turbidities of more than 30 parts per million are high enough to cut off sunshine almost completely except for a shallow layer very close to the surface.

Large streams with long courses, like the Missouri and Mississippi Rivers, are liable to be consistently turbid throughout the year; small streams often show great fluctuations, the peaks coming at times of high run-off and heavy scour. The amount and character of the suspended solids will vary with the geological structure of the catchment

area, and in polluted water courses with the volume and nature of the wastes discharged.

All the material producing turbidity tends to coalesce and to be carried downward by gravity. Turbulence is the strongest factor opposing this tendency. Inorganic particles that are precipitated silt up channels and build bars and shoal areas; the particles remain inert and do not further influence the quality of the water above them. Organic particles form sludge banks that decompose with production of soluble organic and inorganic compounds; these pass into the water. Carbon dioxide and various compounds of nitrogen are here evolved and add to the potential fertility of the stream.

Turbidity may influence stream life in more direct fashion. In the rheoplankton are many animal forms some of which are capable of ingesting and utilizing as food small particles of organic detritus. The sludge of bottom sediments supplies pabulum for worms, larvæ and the larger forms of animal life that do not appear in the flowing water.

Water Movement. — Wind and wave action are not so pronounced in streams as they are in large lakes. Wide and straight stretches of rivers are subjected to a fair amount of surface disturbance which is translated also to the lower depths. Current movements, however, become very significant; eddies, backwaters, pools, rapids, and water-falls present very different environments for aquatic organisms.

Under moderate velocities of flow the life processes of most unicellular plankton organisms continue largely as they do in quiescent water. The filamentous algæ and attached forms are more quickly affected by currents. Swift waters tend to disrupt the filaments and dislodge the sessile organisms. When the flow becomes turbulent the more fragile organisms are destroyed; only the hardier varieties survive. The distribution of velocities in a cross-sectional area of a stream is reflected in the types of growth occurring in different parts of the section. In general the slowly-moving water along the banks harbors somewhat different genera than does the swift body of water in the central channel of the river.

Sedimentation. — The effect of turbidity upon plankton growth has been mentioned in relation to the conditions of light encountered in flowing water. It is in this connection that water movement is of great importance for it is the flow of water that opposes the clarification of turbid streams. The transporting power of streams varies as the sixth power of their velocity and it follows that slight changes in velocity will produce significant effects in the clarification of silt-laden rivers as well as in the formation and movement of sludge deposits and in the shifting

of the banks and bottoms. The carrying power of streams, as expressed by the velocities that will move solid particles on the stream bottom approaches the following values.

TABLE 66
CURRENT VELOCITIES NECESSARY TO MOVE SOLIDS
Metropolitan Sewerage Commission, New York

Kind of Material	Velocity Necessary to Move Along Bottom	
	Feet per Second	Miles per Hour
Fine clay and silt.....	0.25	about $\frac{1}{6}$
Fine sand.....	0.50	" $\frac{1}{3}$
Pebbles half inch in diameter.....	1.0	" $\frac{2}{3}$
Pebbles one inch in diameter.....	2.0	" $1\frac{1}{3}$

When water is perfectly still and the transportation capacity is reduced to zero there are yet other forces, such as buoyancy, friction, and convection currents that oppose sedimentation. The rate at which settling occurs in quiescent water is measured in terms of the hydraulic subsiding value of different materials. It is commonly expressed in mm. per second at 50° F. The hydraulic subsiding value of spherical particles having the specific gravity common to quartz sand (2.65) has been estimated to be as shown in Table 67.

TABLE 67
RATE OF SETTLING IN PURE, STILL WATER
Temperature of Water 50° F.
Specific Gravity of Particles 2.65

Order of Magnitude	Diameter (mm.)	Hydraulic Subsiding Value (mm. per second)	Time Required to Settle One Ft.
Gravel.....	10.	1000	0.3 sec.
Coarse Sand.....	1.	100	3 sec.
Fine Sand.....	0.1	8	38 sec.
Silt.....	0.01	.154	33 min.
Bacteria.....	0.001	.00154	55 hrs.
Clay.....	0.0001	.0000154	230 days
Colloidal Particles..	0.00001	.000000154	63 years

It is evident that particles finer than silt will ordinarily not precipitate even in quiet water. The rate of settling decreases as the specific gravity of the particles decreases, as the current velocity increases, and as the water becomes colder or denser. It is changed viscosity, however, rather than changed density that affects the rate of settling.

Bottom Sediments. — No study of the quality of stream waters is complete without a knowledge of the sediments deposited on the bed of the streams. Floods, droughts, and the fluctuating character and volume of wastes produce wide extremes of quality in the water. Only repeated analyses over a considerable period of time enable deductions to be made as to average quality. Bottom sediments, however, are built up from material carried at different times by the water and thrown down over relatively long periods of time. They are composites of suspended organic and inorganic impurities; they even contain to some extent portions of the plankton growth, more particularly members of the immotile plankton species. In addition these deposits are often teeming with larger animal forms of life that are busily engaged in digesting the accumulation of organic material, a most important function in the purification of stream waters. Were it not for their activities stream beds would become choked with organic débris and foul conditions would ensue.

Bottom sediments, then, reflect the average condition of the water flowing over them, oftentimes more perfectly than do analyses. Examination of their physical properties, color, odor and consistency and of their biological content introduces summarized evidence of the stream's behavior over a considerable period of time. Such an examination goes hand in hand with studies of the rheoplankton and environmental influences.

Hydrography. — Comparing the hydrography of rivers with that of lakes our attention is first focused on the greater shore line development existing in streams. The absolute development of shore line in Lake St. Clair for example determined by the method of Seligo (see page 259) is given by Reighard as 9.23 while Kofoid estimated that of the Illinois River from Utica to the mouth as approximately 17.1 at high water and 78.3 at low water. In the shoals near the river banks currents are sluggish, larger aquatic vegetation flourishes, ground water enters the stream and swamps abound. The diversity of the littoral environment is reflected in a greater variety of microscopic organisms here existent than in the channel proper. The types of life found differ with the nature of the shore. Springs that during the summer carry colder water into the streams favor the development of diatoms. Swamps and back waters permit the growth of grass-green and blue-green algae.

Larger aquatic vegetation offers anchorage for attached forms and shelter for protozoa, but it also deprives the plankton of much food material.

Floods and Droughts. — The instability of the environment of the rheoplankton is exemplified perhaps most by the effects wrought by variations in the river stage. These result in sudden changes in depth, area, volume, strength of current, quality of water, character of bottom, and many other factors. Rising stages are most disastrous to the content of river plankton. Flood waters are generally more turbid, carry less food material, and often reach destructive velocities. The chief cause for reduction in plankton content, however, is dilution, i.e., the mixing of old plankton-rich waters with new plankton-poor ones. The effects upon backwaters are as marked as upon the channel itself.

Kofoed reports the action of the December, 1895, flood on the Illinois River as follows:

Rising suddenly from low levels (3 ft.) to overflow stage (12.6 ft.) in 12 days, it depleted the channel plankton from 2.6 cc. per m.³ in the initial stages to .08 on the 25th, if not, indeed, earlier. Not only did it thus depress the plankton content in channel waters but, with a less catastrophic completeness, that also in the back waters. Thus in Thompson's Lake the plankton fell from 1.87 cc. on the 19th to .13 on the 28th; in Quiver and Dogfish lakes, from .63 and 10.57 to .29 and .06, and in Flag Lake, from 6.38 to 3.26. The effectiveness of the depletion was greatest where overflow currents were best established, as in Thompson's and Dogfish lakes, and least where the currents were slight and impounding greatest, as in Flag Lake.

Falling stages in general present the opposite picture. They are less sudden in character and bring with them a gradual reconstruction of the flora and fauna of the waters.

River Sampling. — The changing hydrographic character of streams, varying from place to place and fluctuating from day to day introduces the question of how to collect representative samples. When a single sample is to be taken or an integrated vertical sample it is commonly taken in midstream. When several collections are to be made their logical distribution calls for sampling points so located that equal amounts of water pass through the different areas at whose center of mass the samples are taken. While this is theoretically possible, practical considerations demand a simpler approach.

In the Ohio River studies the United States Public Health Service selected three sampling points as follows: the cross-section below an average elevation of water surface was divided vertically into three equal areas and integrated samples were taken along the vertical dividing each area into two equal parts. For gage heights other than the average the sampling stations were not shifted horizontally. Chemical

and bacteriological samples were taken at mid-depth and the sampling point was adjusted vertically to each gage height. The method of locating the sampling stations is illustrated in Fig. 82.

The inclined haul method of sampling is particularly useful in river work. By it a sample integrated both horizontally and vertically can be obtained. Although developed for use in connection with the plankton net it is possible to employ it also with other sampling devices.

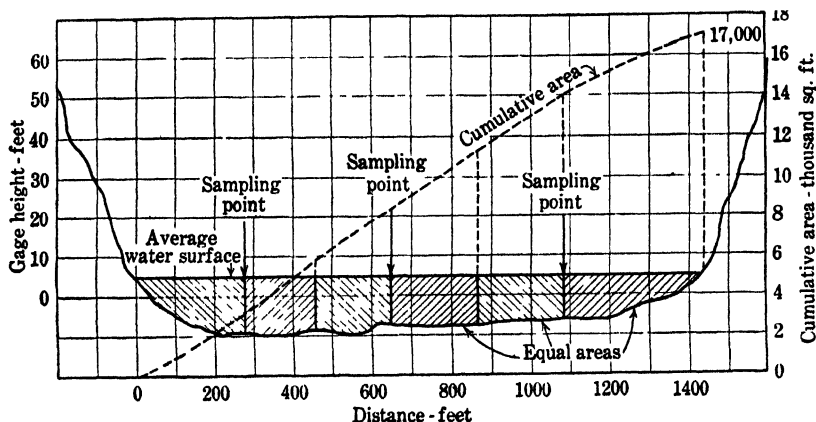


FIG. 82. — Method of Locating Sampling Points on Stream Cross-Section.
After Frost; U. S. Public Health Bulletin 143.

CHEMICAL CONDITIONS

Data presented elsewhere in this chapter suffice to show that the aggregation of microscopic life in streams is often large but commonly falls short of that found in lakes and reservoirs. The occurrence of these living forms is fundamentally dependent upon food; so we must expect to find the chemical environment in streams similar in many respects to that in lakes and ponds. At least, conditions must be comparable in a qualitative way, for we cannot imagine that the nutrition of similar cells will be satisfied in running water in a totally different manner from that obtained in standing water. The rheoplankton, as compared with the limnoplankton, is, however, subjected to diminished constancy of needed substances through the changing character of streams in their courses, and to greater irregularity in the concentration of these substances that results from the entrance of waste discharges and from fluctuating volumes of flow. These frequent changes, together with the influences exerted in streams by various physical conditions, already discussed, make for extreme instability of the fluvial environment.

Dissolved Oxygen. — All aquatic organisms demand oxygen to satisfy the needs of respiration. With few exceptions this demand, however small in amount, must be met by *dissolved oxygen*. There is, consequently, a depletion of the initial store of dissolved oxygen due to biological activity. Also, a certain loss of oxygen occurs through chemical oxidation of organic matter. Ordinarily this is small but it increases with the concentration of fresh organic material. There is compensation for these losses in two ways: (1) the phytoplankton and submerged vegetation release oxygen through photosynthesis at a much higher rate than they consume it through respiration, (2) contact of the water surface with the air results in solution of atmospheric oxygen and its diffusion through the liquid. Thus through losses and through compensation of losses there are changes in the oxygen content of streams which operate in two ways, first, to affect the living organisms and, second, to affect the chemical environment. This dual aspect is common to many observed changes in the chemical constituents of streams.

Reaëration by Heliophilous Organisms. — The extent to which the dissolved oxygen content of lakes and ponds may be increased by activity of the phytoplankton and larger plants has been pointed out in Chapter VIII. In flowing streams such production is usually not as important a factor in maintaining the presence of oxygen as is the solution of atmospheric oxygen. Turbulence and other physical conditions do not favor heavy growths of these heliophilous organisms.

Streams whose course is flanked with large stretches of shallow water and submerged flatland, where the flow is sluggish, may, on the other hand, derive considerable amounts of oxygen from photosynthetic processes that go forward in these quiet waters. Decreased turbidity, relative quiescence, and proximity to bottom sources of food favor aquatic growth; the emerging water consequently may be more highly oxygenated than that entering from the channel. If the areas of shallow flow are relatively large, then plant forms of life exercise an important influence on the economy of the river by aiding to maintain an aërobic environment.

Polluted tidal streams afford the best examples of reaëration from flatlands, for the oxygen production of the latter is daily flushed by ebbing tides into the streams where decomposition has resulted in a reduction of dissolved oxygen. Purdy in his study of Potomac River flats gave considerable attention to this matter. Table 68 presents a summary of his work. The samples were collected between September 3 and November 12, a period when growth was not at maximum. During the latter part of November and again in the early spring there was little difference

between oxygen values in the channel and on the flats, the flats water exhibiting a slightly lower saturation value. This indicated a lower rate of oxygen production and relatively greater bacterial activity.

TABLE 68
DISSOLVED OXYGEN ON FLATS AND IN THE CHANNEL OF THE POTOMAC RIVER
After Purdy

Sampling Point	Number of Samples	Dissolved Oxygen Content, Per Cent of Saturation		
		Highest	Lowest	Average
Flats.....	27	128	57	87
River channel.....	12	102	50	71

Purdy likens the movements of tidewater over flatlands to movements of air in the lungs. "Ordinary tides correspond to ordinary breathing — a very high tide is analogous to a deep inhalation of air and a very low tide is similar to an enforced exhalation. Finally the 'residual air' in the lungs typifies the large body of 'residual' water which remains in the flats, continually renewed, constantly depleted, yet never exhausted."

The effect of these tidal changes is like that of brief impounding of water. Current velocities are lowered, sedimentation takes place, being greatly aided by the enormous area of exposed plant surfaces, and reaeration is promoted by contact with the air and by photosynthesis. Physical, chemical and biological changes manifest themselves.

Sunlight and Oxygen Production. — Inasmuch as photosynthesis and the production of oxygen can take place only in the presence of light it is reasonable to expect that the extent to which they manifest their effects will vary with the intensity of the light. Other things being equal sunny days will produce the maximum concentration of oxygen; days with heavily clouded skies will produce the minimum. A statement of prevailing meteorological conditions should, therefore, accompany the report of oxygen findings if the water contains numerous heliophilous organisms. Likewise, the observations of any one day will not represent average conditions if the day is one of continuous sunshine or one of uniformly cloudy conditions.

In his study of the Potomac River flats, Purdy obtained strikingly different oxygen values on sunny and on cloudy days. In these broad

shallow areas which favored the development of microscopic and larger forms of plant life oxygen was frequently in excess of saturation values. Table 69 presents a summary of these observations.

TABLE 69
OXYGEN IN WATER OF POTOMAC RIVER FLATS ON SUNNY AND CLOUDY DAYS
After Purdy

Sky Conditions	Number of Samples	Dissolved Oxygen Content, Per Cent of Saturation		
		Highest	Lowest	Average
Sunny.....	16	128	72	99
Cloudy, overcast....	11	87	57	69

The average oxygen concentration in the river channel at stations where water from the flats entered the stream at ebb tide was also higher on sunny days. In the upper part of the stream, where the channel was narrow and flats did not exist, the average concentration was the same on sunny and cloudy days. Plankton life at this point was not abundant.

Reaëration by Absorption. — By far the most important source of oxygen in streams is the air in contact with the water surface. The rate at which it is absorbed depends upon factors which have been cited on p. 190, chief of which is the degree of under-saturation, or saturation deficit, of the water. If other factors than diffusion did not also condition the rate of absorption there would not be in heavily polluted streams enough oxygen to carry forward the processes of purification through a very great depth of water. Diffusion of oxygen in quiet water is a slow process, but where there is turbulence produced by irregularities in the stream bed, or by wave action, diffusion gains acceleration. This greatly increases the rapidity with which oxygen is carried to depths. This is accomplished by bringing small volumes of water high in oxygen in contact with volumes that are low in oxygen. The difference in concentration increases the rate of diffusion. Turbulence also promotes dissemination of the gas through mechanical mixing and through greater streaming effects. The latter result from surface evaporation which increases density and sets up downward currents. The rate of oxygen diffusion, and, therefore, absorption is higher in streams than in standing bodies of water by virtue of the greater turbulence of the former.

Reaëration is modified in the stream by all kinds of biochemical activities that call for oxygen, that is by the biochemical oxygen demand. The higher the rate of biochemical oxygen demand the longer becomes the time for a given deficit to be replenished and for the oxygen to approach the saturation point.

Vertical Variations. — It has been pointed out previously that thermal stratification in the channels of streams is not marked owing to the mixing of water that is induced by turbulence of flow. Where sluggish velocities allow thermal differences to be established these are liable to sudden effacement by floods and rapid changes in air temperature. The general mixing of top and bottom water also operates to maintain fairly uniform concentrations of dissolved oxygen and to mask the more rapid consumption of oxygen that takes place at the bottom.

During midsummer some evidence of thermal stratification may be noted in streams when turbulence is slight and the depth is considerable. Figure 81 presents such a condition that existed in the Genesee River at Rochester, N. Y., on July 15, 1912. The surface temperature was 80° and the bottom 70° F. In Fig. 83 the corresponding values for oxygen saturation are given. For over three miles the surface saturation was between 30 and 40 per cent and the bottom saturation between depths of 16 and 30 feet was less than 10 per cent. The whole distance represented a zone of decomposition but the greatest activity was displayed in the bottom water. The diagram shows the rapidity of longitudinal changes in oxygen values near the mouth of the river, particularly in the bottom water. Admixture of the cleaner water of Lake Ontario was largely responsible for these. The example of a heavily polluted stream is chosen, because it illustrates the point in question better than a clean stream could.

Another diagram might be presented to show that on July 24 thermal stratification was practically absent and that through most of the distance covered by Fig. 83 the water contained uniformly less than 10 per cent of oxygen from surface to bottom. Diurnal changes in air temperature induced vertical circulation of water. The factor of turbulence was of less consequence on the latter date than on the former, the discharge of the stream on July 24 being 346 cubic feet per second, whereas on July 15 it was 500 cubic feet per second.

Longitudinal and Horizontal Variations. — More striking by far than the vertical differences in oxygen concentration are the longitudinal differences. These are caused by the forces of self-purification, by discharge into the stream of decomposable wastes and by the acquisition of considerable volumes of relatively clean water from other streams, from flats and from the ground. A stream receiving a heavy burden

of pollution will run the gamut of oxygen values, and if there are repeated acquisitions of polluting wastes these values will rise and fall in recurrent pulses throughout the length of the stream. The causes and effects of longitudinal changes in dissolved oxygen in polluted streams are discussed at length in Chapter XII.

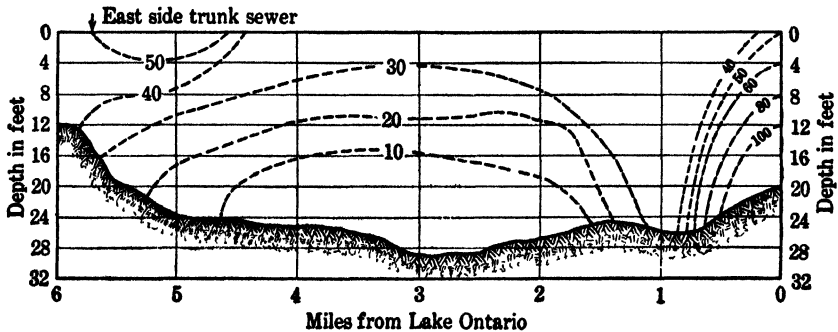


FIG. 83. — Percentage Saturation of Dissolved Oxygen. Genesee River at Rochester, N. Y., July 15, 1912.

Horizontal distribution of oxygen is apt to be uniform at given depths. Exceptions to this general condition are sometimes noted where extensive backwater areas are produced by eddies, and more often below the entrance of tributary streams. In the latter case the character of the channel may induce incoming water to keep close to one bank or the other for a considerable distance.

Carbon Dioxide. — The amount of free carbon dioxide found in stream water is largely the resultant of the same processes that influence the dissolved oxygen, but changes are in the opposite direction to those of oxygen. Photosynthesis produces oxygen, but at the expense of carbon dioxide, the carbon of which goes into living substance. Respiration consumes oxygen and carbon is released from the cell in the form of carbon dioxide. Bacterial decomposition of carbonaceous material tends ultimately to oxidation of the carbon atom with removal of oxygen from solution and the formation of soluble carbon dioxide. The last-named process gives rise to enormous quantities of this gas. A ton of carbon so oxidized yields over three million grams of carbon dioxide, the equivalent of 30 parts per million in 100 million liters or 26 million gallons of water. In this way Nature restores for the use of subsequent plant growth the carbon that was locked up in dead cell substance.

The atmosphere affords an inexhaustible supply of carbon dioxide, but this is not absorbed unless the concentration of this gas in the water falls to low values, below 1 or 2 parts per million. In fact there is the

opposite tendency in polluted streams, namely, for the gas to pass into the air and so to establish equilibrium with the atmospheric partial pressure.

Ground waters may carry in considerable quantities of CO_2 and add to the total amount present when the concentration in the stream is less than that of the ground water. Seepage of water from the ground can frequently be marked along stream courses by local growths of attached algae and larger plants that are favored by the higher concentration of carbon dioxide.

At those seasons when submerged vegetation is decaying there is a constant contribution of carbon dioxide from flats and areas of shallow flow, but this is not large in amount. Purdy found that the tide water from the Potomac River flats during November and again in the spring averaged between 2 and 3 parts per million of carbon dioxide, about the same as the channel water. Evidently plant decay takes place gradually. Its effects may also be marked by the growths of algae that are found to flourish after the growing period for larger vegetation has passed. Swamps and marshes that are not subject to tidal action may discharge into streams large amounts of carbon dioxide for short periods of time. These coincide with floods and times of high stream flow.

Carbon Dioxide and Plankton Growth. — Streams undefiled by any other than Nature's pollution will contain at most only a few parts per million of carbon dioxide. This is one of several reasons why they fail to develop large crops of plankton life. Pollution with heavy organic wastes, such as sewage, completely alters conditions. Oxidation of carbon in the flowing water and in bottom deposits produces great volumes of carbon dioxide in the zones of degradation and decomposition; this and other food substances increase the potential fertility of the water. The result is a marked stimulus to development of the phytoplankton whenever other environmental factors are suitable. Usually before such time arrives there is a large loss of carbon dioxide to the air. If this loss were prevented the limits of plankton growth would in many streams far exceed those that are observed.

Mineral Constituents. — Analysis does not indicate mineral substances in the water of normal streams that are different to those found in lakes and ponds. As in the latter, the quantities will vary widely, largely owing to the diversified geology of different regions. Lakes produce equalizing effects, however, that are lacking in most streams and so exhibit narrower fluctuations in the amount of mineral substances than do streams; the waters of the latter are subject to dilution by floods and concentration by droughts. Flood waters commonly contain smaller amounts of such salts as bicarbonates, sulphates and

chlorides than the streams into which they run; this by reason of their shorter period of contact with the soil and underlying strata. Hence the usual condition that minimum values for these substances are obtained at seasons of high flow and maximum values at times of low flow. When streams are low the infiltration of ground water is often the controlling cause of higher mineralization. Ground water flow is more constant than that of surface water and brings with it a content of dissolved mineral matter that is usually higher than that normally found in the stream.

The most important mineral compounds from the standpoint of availability for growth of the hydrophytes are ammonium salts and nitrates, to be mentioned later, and the bicarbonates and silica. The latter is utilized by the largest group among the phytoplankton, the diatomaceæ.

Influence of Pollution. — The common practice of using streams for the conveyance and disposal of human and industrial wastes in many instances materially alters the content of mineral constituents. Sewage, for example, contains a large amount of chlorides that appears unchanged in the water. It is also relatively high in bicarbonates and sulphates and contains considerable iron and magnesium. The organic nitrogen of the sewage is ultimately converted to nitrites and nitrates, that, downstream, add to the amount of these mineral substances already present in the water.

Trades' waste very often contributes calcium and magnesium compounds in large amounts. Caustic lime precipitates the natural bicarbonate alkalinity of the stream as carbonate and builds up deposits on the stream bed. Sulphites neutralize the alkalinity and produce incrustants. Salt water discharges from the oil and salt industries add neutral salts, principally the chlorides of sodium and magnesium. With the exception of the nitrates none of the substances mentioned exerts any particular influence upon river plankton, except as it may exclude light by formation of precipitates or prove actually toxic in highly concentrated doses.

Acid Waters. — Acid wastes from iron pickling processes and drainage from mines are discharged into some streams of the country in large volumes and profoundly affect chemical and biological conditions. Their acidity is primarily due to sulphuric acid; sulphates of iron accompany the acid. Carbonate hardness of streams is converted to the non-carbonate form (sulphates) by such wastes, carbon dioxide being liberated by the reaction. Dissolved iron salts increase the soap hardness, as does any excess of acid in the water. Oxidation and admixture with alkaline water precipitate the iron which in turn causes turbid-

ity and increases bottom deposits. The Monongahela River which unites with the Allegheny at Pittsburgh to form the Ohio is a striking example of pollution by both pickling liquors and mine waters. It is normally acid to methyl orange in its lower reaches.

Acid pollution brings about great biological changes. Purdy in his studies of the Ohio River Basin calls attention to the strong sterilizing action of the acid wastes in the Monongahela and the resulting low bacterial content. Unusual conditions were also noted in the plankton. Although this river receives a great deal of sewage and affords by its slack water basins ample time for digestion of organic matter it manifests extreme poverty in content of phytoplankton, which are practically absent. Ciliated protozoa are also absent. The larger plankton forms are very numerous but the variety is confined to a few genera of the rotatoria. The free mineral acid apparently acts directly and prejudicially upon the cells of the phytoplankton. The protozoa may be affected in similar manner, or may find scanty forage in the small number of bacteria.

Ammonia Nitrogen. — Of all the inorganic compounds found in the water of streams those of nitrogen, particularly ammonium salts and nitrates, are of greatest influence upon the production of river plankton. Nitrites, intermediate between the two in their state of oxidation, are usually found in smaller concentration. They do not accumulate in large amounts, being further oxidized to nitrates, and show little correlation with plankton growths. Ammonium salts, commonly designated as "ammonia nitrogen" or "free ammonia" represent the first stage in the mineralization of organic nitrogen. All streams contain at least traces of ammonia nitrogen; those laden with the extracted products of dead vegetation contain moderate amounts; streams defiled with organic wastes from human habitations and industry contain more than either of the others, the amount depending upon the concentration of waste and the time elapsing after its advent into the stream.

In the presence of dissolved oxygen ammonium salts do not tend to accumulate, being oxidized by nitrifying bacteria to more stable compounds. At the same time they are assimilated by plant life as a source of nitrogen. The highest values for ammonia nitrogen are obtained in the presence of large amounts of decomposing organic matter of which it is an index.

In spite of its availability as food for large and small forms of plant life high ammonia nitrogen in stream waters does not often afford certain evidence of an abundance of such organisms. Other factors preclude their development; turbidity excludes light, there may be scarcity of oxygen, and time for breeding may not be allowed before

the ammonium compounds are oxidized. Potential, rather than immediate, ability to develop plant life is, therefore, more often indicated by high ammonia nitrogen.

Nitrates. — Nitrogen in its final oxidized form as nitrate is never great in amount except in those waters that have received human or animal pollution at a relatively distant time in the past. There is one modification of this statement to be made, the fact that comparatively high values for nitrate nitrogen are observed below large areas of cultivated land. In such cases nitrates are dissolved from the soil, where they have been built up from the nitrogen of the air and to a larger extent from the nitrogen in organic animal wastes used as fertilizer. It is true that a considerable period of time has elapsed during this transformation, but the actual entrance of the nitrogen into a stream that is under examination may have occurred very recently. Under such conditions the nitrates may be accompanied by considerable soil pollution that has not been oxidized.

It is not uncommon to find in streams that have recovered through natural processes from heavy pollution several parts per million of nitrogen in the form of nitrates. Here is evidence of immediate fertility, for according to present knowledge nitrates represent the most easily assimilated form of nitrogen that is available for plant growth. Other environmental conditions must be favorable, however, to permit of nitrate utilization, and time must intervene for growth and reproduction before a heavy crop of aquatic life is brought forth.

It is not necessary that large amounts of nitrate be present in order to assure productiveness. If the input is constant, and large enough to maintain an excess over the ability for growth, the excess need not be large in order to assure the development of phytoplankton and larger plants. Some waters convey in the aggregate huge amounts of nitrates built up from sewage. These may be produced by oxidation of organic nitrogen within the stream or they may be contributed directly by effluents of sewage works. As an example, a stable effluent with a content of 5 parts per million of nitrate nitrogen, when discharged at the rate of 10 million gallons per day, will add 415 pounds of nitrate nitrogen daily to any water receiving it. This is at the rate of over 12,000 pounds, or 6 tons per month, the equivalent of 36 tons of sodium nitrate. Such a tonnage, if applied to the soil, would stimulate the growth of enormous land crops. Added to water, even though the dilution be very great, there yet remains a constantly available nitrogen content sufficient to bring forth a luxuriant aquatic crop.

In using nitrate nitrogen as an indicator of fertility toward plants it should be borne in mind that the nitrogen found represents only that

which has not been used, not the total produced. That which has been assimilated by growing plants has been removed from the realm of mineral analysis and appears as organic nitrogen.

All things considered, the amount of nitrates found in a stream cannot be said to indicate the extent of the plankton population or the development of larger aquatic plants. It does provide, however, an inventory of the most available form of nitrogen. Thus Fig. 84 shows that in the Illinois River the greatest plankton production was not coincident with the periods of highest nitrate values but followed them. The peak of plankton development corresponded to the periods of lowest nitrate values, a manifestation of cause and effect.

Organic Matter. — In moving waters most of the organic matter is inert and of comparatively recent origin; it is accompanied by large numbers of bacteria because of its easy availability as food, and is, therefore, subject to rapid putrefaction and decay. From the chemical standpoint plankton life also forms a part of the organic content but it is not subject to disintegrating changes until after death.

Sources of Organic Matter. — The sources of organic material are the erosion of the soil, extraction of deposits of dead animal and plant substance and the contribution of waste from human habitations and industry. Such complexity of origin brings in every class of nature's food substances, fats, carbohydrates, proteids and in addition their derivatives that have resulted from bacterial and animal metabolism. Some are soluble, others are carried in suspension. The task of digesting these multifarious compounds and of returning their elements through proper cycles to Nature's use is one that calls for employment of a variety of forces. The latter are discussed under Stream Purification in Chapter XII.

Limitations of Analysis. — Chemical studies of organic matter in water are exceedingly involved and, as ordinarily conducted, do not identify specific compounds. Neither do they distinguish between living and lifeless material. Certain tests directly record the total amount of the composite mixtures, such as the determination of volatile solids, or the amount of some one element in these mixtures, such as the determinations of organic nitrogen and albuminoid nitrogen. Other tests, like those of oxygen consumed and biochemical oxygen demand, by inference show the amount of organic matter that possesses certain properties, — in these tests the tendency to be oxidized. On the whole, we obtain by analysis a very imperfect picture of the chemical composition of organic matter in natural waters. Some of the more recent attempts to extend this information, which are suggestive of a greater knowledge to come, have been described in Chapter VIII.

Cyclic Changes in Organic Matter. — A long stream course is a vast laboratory equipped for many processes. Into it is carried every form of waste. Here through the seasons and the years nature rings the changes on varied conditions, but always with the purpose that the experiment shall end with digestion of the raw products. At one time there is high dilution, at another there is heavy concentration; now there is turbulence, sunshine, heat or evidence of teeming life, then comes quiescence, darkness, cold or restricted life. These and other factors, some of which are controlled by acts of man, determine whether the resultant processes will carry forward the aim of the experiment or retard it.

In a flowing stream one may best observe the progress of the cycles through which pass those elements that are most important to the living cell. Carbon, a constituent of all organic matter emerges from the processes of putrefaction and decay in the form of carbon dioxide and is assimilated by plant life to form carbohydrates and protein. The plants become the food of small animal life that falls prey to larger animal forms; fats are an added product. The products of metabolism and the final death of both plants and animals complete the cycle and carbon again appears in inert form. Short circuits appear in this typical cycle; respiration produces carbon dioxide without the intermediate stages of death and decay, and there is direct utilization of decomposing waste by certain low forms of animal life.

Sulphur, found in the complex structure of dead cells, also passes through a cycle, putrefaction forming hydrogen sulphide. Oxidation produces sulphates; plant life utilizes these to form its protein, the latter appearing later in the animal cell. Death of plants and animals returns the sulphur to the first stage of the cycle.

The nitrogen cycle is more complex than either of the others. Putrefaction and decay of dead organisms and waste materials give rise to ammonia compounds that are assimilated by some plant organisms; usually the nitrogen of the ammonia compounds is carried to nitrites and nitrates by oxidizing bacteria. The oxidized nitrogen becomes one of the chief foods of plants in building up protein. To a slight extent oxidation is sometimes reversed and nitrates are reduced to nitrogen that may be dissolved in the water or escape to the air, thus representing a loss. Plant protein becomes animal protein. Finally death of both plants and animals returns nitrogen to the processes of putrefaction and decay. A short circuit in the cycle is the digestion of protein with elimination of urea, from which ammonia is derived without putrefaction.

Organic Food of the Plankton. — Ultimate mineralization after passing through the course of the cycles described is not the only method by

which organic waste is digested. River plankton are a powerful force in this direction, as are other low forms of life, such as the worms and larvæ. The richness of the fluviatile environment in organic substances and bacteria provides a fertile field for development of animal life and leads to conversion of much of this substance into living material.

The phytoplankton is also of importance in this rôle. Marsson has pointed out the ability of some of the microscopic plant forms to obtain their food, not alone by assimilation of mineral substances but, also, by absorption of albumins and similar nitrogenous compounds. Further, "Subsequent physiological experiments showed that diatoms, as well as green algæ, can not only absorb carbon compounds from dissolved organic material, but also nitrogen; and that when carbon dioxide is rigidly excluded from the water they can digest diluted volatile fatty acids, amido acids, skatol, urea, peptone and other substances . . ."

"This direct absorption of dissolved organic matter by algæ is of the utmost importance for the purification of streams by eliminating therefrom the soluble products of putrefaction. As the algæ are busily engaged in the work of producing fresh albumen, starch and indirectly fats, from animal and vegetable matter, it follows naturally that numerous animal organisms will develop in the water to make use of these products and convert them into living flesh."

Chemical Analysis and Plankton Growth. — When data from all the elements of a chemical analysis are available for consideration much information is at hand concerning the capacity of a stream to support plankton life. Certain important food elements, as carbon dioxide and nitrates are indicated; the presence or absence of dissolved oxygen is shown; nitrogen is identified in its different stages of oxidation. Analysis does not disclose the amount of organic nitrogen immediately available as food, nor does it differentiate between nitrogen in such food material and that in the living cells of the plankton.

The different nitrogen tests are the most valuable ones in a chemical analysis for judging plankton capacity of streams, but, unfortunately, it is difficult to obtain accurate correlations between them and actual plankton numbers. Repeated examinations on the same stream will disclose certain general trends in the upward and downward movements of the nitrogen values that are coincident with changes in plankton population. However, chemical analysis does not evaluate the effect of physical and biological forces that are at work; consequently prediction of plankton findings from chemical data alone is a precarious undertaking.

BIOLOGICAL CONDITIONS

The use of the term rheoplankton seems to imply that the microscopic organisms found in streams are different from those that dwell in standing waters. This, as Kofoid has pointed out, is not so. Summarizing his investigations of the Illinois River he says that the river plankton is distinguished from the lake plankton by the following characters:

1. It is a polymixic plankton. This is due to the mingling of planktons from all sources in the drainage basin, especially from tributary backwaters, and the consequent seeding of the channel waters with a great range and variety of organisms. In all of our collections in channel waters monotonous planktons can scarcely be said to be present. The nearest approach to such conditions occurred at low water stages when channel waters are most fully isolated.

2. It is subject to extreme fluctuations in quantity and constitution. This naturally follows from the manifold factors of the fluvial environment and the directness with which they impinge upon the plankton. Changes in volume, contact of shore and bottom, access of heat and light and changes in chemical constituents are frequently both more extensive and more widely effective in the stream than they are in the other types of aquatic environment. In consequence, the plankton of the stream is subject to more catastrophic changes than that of the lake.

3. The potamoplankton is not characterized by any species peculiar to it, nor by any precise assemblages of eulimnetic organisms. It may be distinguished, in a general way only, by the greater proportion of littoral or benthal forms which are mingled with the more typical planktons.

Constituent Groups of the River Plankton. — The great variety of the river plankton pointed out by Kofoid is illustrated by an examination of the analyses of river catches such as were obtained in the Illinois and San Joaquin rivers. Table 70 will serve to indicate in some measure the proportionate representation of the most typical constituent groups.

For a comparison of the figures in this table it is well to state that many of the Illinois catches were filter-paper determinations whereas all of the San Joaquin collections were made by nets. The relative presence of different groups is of great interest. The predominance of the plant forms over the animal forms is especially noteworthy. The ratio of phytoplankton to zoöplankton as we shall see in the next chapter in a measure reflects the status of stream pollution. In our example this ratio in the polluted Illinois River is about 5 to 1, in the relatively clean San Joaquin River almost 10 to 1. Comparisons of the synthetic and analytic organisms yield similar ratios, namely 18 to 1 and 25 to 1, respectively.

Production of Rheoplankton and Limnoplankton Compared. — The annual production of microscopic organisms in rivers is generally not as great as that in lakes. In the Illinois River during the years 1894 to

TABLE 70

MAJOR CONSTITUENT GROUPS OF THE RHEOPLANKTON
Average Catches — Illinois River, 1898; San Joaquin River, 1913

	Illinois River		San Joaquin River	
	Number of Forms	Number of Individuals per m. ³	Number of Forms	Number of Individuals per m. ³
Schizomycetes.....	3	57,142,822*	1	19,905
Algae:				
Cyanophyceæ.....	9	85,909,985	18	1,292,380
Chlorophyceæ.....	33	53,175,105	13	1,499,479
Diatomaceæ.....	29	396,192,716	59	39,478,317
Conjugatæ.....	7	48,459	9	331,687
Phanerogamia:.....	2	9
Total phytoplankton.....	83	535,326,274	100	42,622,068
Protozoa — total.....	185	111,731,000	53	3,494,065
Mastigophora.....	68	95,856,449	20	2,629,112
Rhizopoda.....	59	55,364	9	150,817
Heliozoa.....	5	4,871	5	257,232
Sporozoa.....	3	1,638
Ciliata.....	45	15,812,346	16	447,909
Suctorior.....	5	332	3	8,995
Rotifera.....	104	592,416	43	883,510
Entomostraca — total.....	43	47,041	7	22,206
Cladocera.....	26	6,242	4	7,384
Ostracoda.....	4	191
Copepoda.....	13	40,608	3	14,822
Miscellaneous.....	114	9,393	7	10,449
Total zoöplankton.....	446	112,379,850	110	4,410,230
Total plankton enumerated.....	529	647,706,124	210	47,032,298
Synthetic (chlorophyll-bearing).		613,017,986		45,231,275
Analytic (non-chlorophyll-bearing).....		34,687,781		1,801,023

* Not included in the totals.

1899 the ratio of channel plankton to backwater plankton was 1 to 3.4. During the colder months of the year river organisms are particularly scarce. Hydrologists have called this the replenishing and storage period of lakes and reservoirs. In this time, therefore, the backwaters retain their microscopic flora and fauna while in the seasons of high run-off they unload their living burden and the differences in the catch are equalized. The seasonal ratios found in the Illinois River are shown in Table 71.

TABLE 71
 RATIO OF CHANNEL PLANKTON TO BACKWATER PLANKTON
 Illinois River, 1894 to 1899
After Kofoid

Month	Ratio	Month	Ratio
January.....	1 to 8	July.....	1 to 0.9 (1.3)*
February.....	1 to 9	August.....	1 to 3.6
March.....	1 to 9	September.....	1 to 5
April.....	1 to 2.5	October.....	1 to 8.7
May.....	1 to 3	November.....	1 to 17
June.....	1 to 0.9 (4.4)*	December.....	1 to 9.6

* Omitting 1895.

It is evident that the channel plankton of the Illinois River in a large degree has its source in the impounded backwaters of the river system. This does not mean, however, that development of organisms does not take place in the river itself. Reproduction continues in the river channel and the composition of the plankton is varied in accordance with the changed conditions of existence. The growth of organisms in channels seeded by backwaters depends largely upon the time during which they stay in their swiftly moving habitat. The longer the time the greater will be the production of an indigenous or typical rheoplankton.

Seasonal Distribution of the Rheoplankton. — The seasonal distribution of river organisms is varied from that of lake dwellers largely by the interference of the fluctuating hydrographic conditions of river basins. This is indicated in Figs. 84 and 85. Figure 84 shows the seasonal distribution of total plankton and diatoms plotted against the thermograph and hydrograph of the Illinois River at Havana. The correlation between river stages and microscopic organisms although not very high is suggestive. In Fig. 85 is shown the seasonal variation of plankton in the San Joaquin River at Stockton, California. Here flood conditions persist from December to July and low water obtains during the remaining months. Plankton catches in this case go counter to the hydrograph. It is readily seen that a knowledge of hydrographic characteristics is essential to a correct interpretation of river catches. Other factors affecting seasonal variations are very similar to those operative in lakes and are not in need of an extensive commentary.

A study of Figs. 84 and 85 shows that plankton production occurs

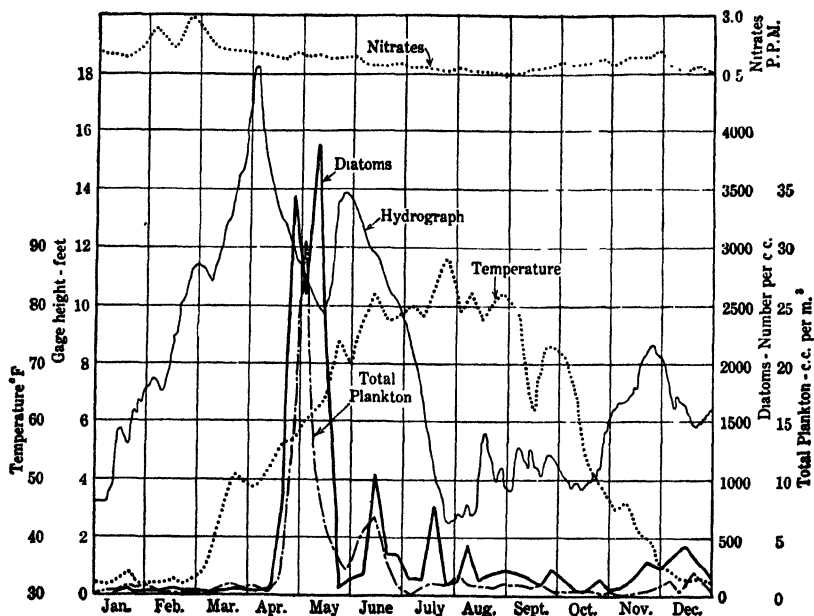


FIG. 84. — Seasonal Distribution of Total Plankton and Diatoms Compared with the Thermograph and Hydrograph of the Illinois River at Havana, 1898. *After Kofoid.*

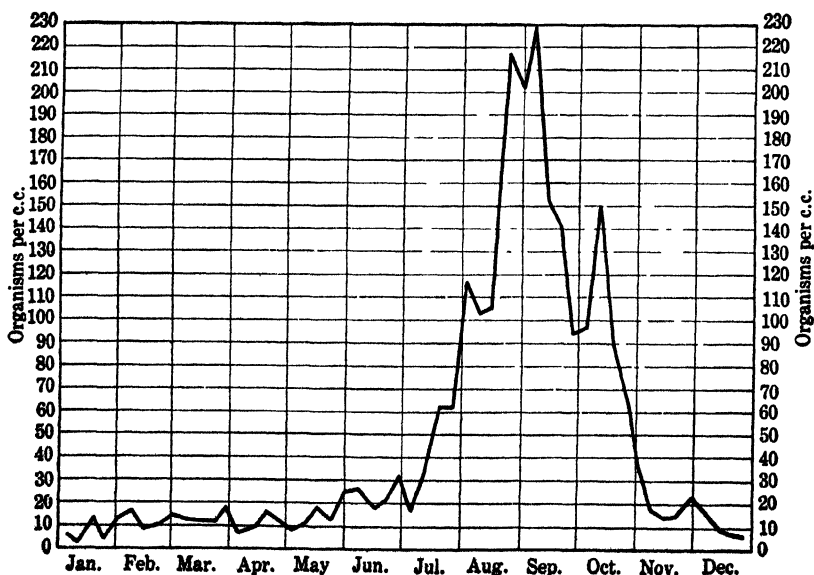


FIG. 85. — Seasonal Distribution of Total Plankton. San Joaquin River, 1913. *After Allen.*

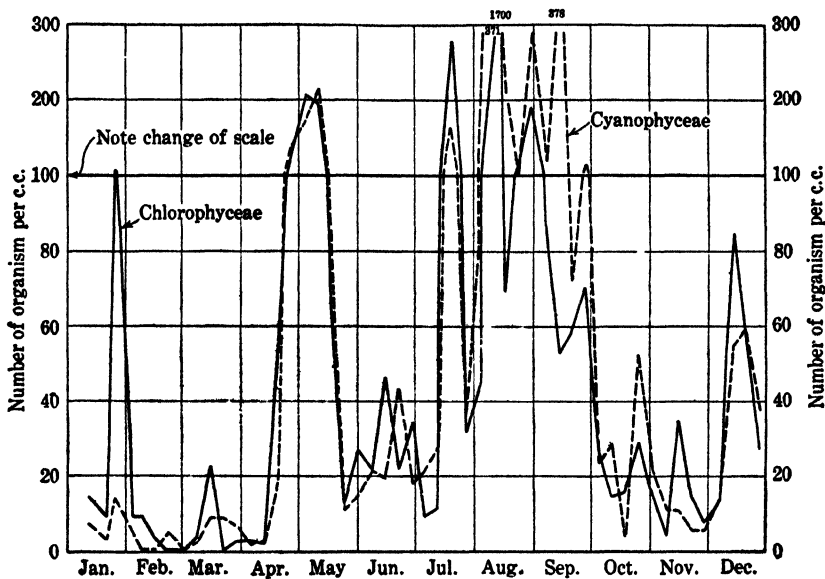


FIG. 86. — Seasonal Distribution of Chlorophyceae and Cyanophyceae. Illinois River at Havana, 1898. After Kofoid.

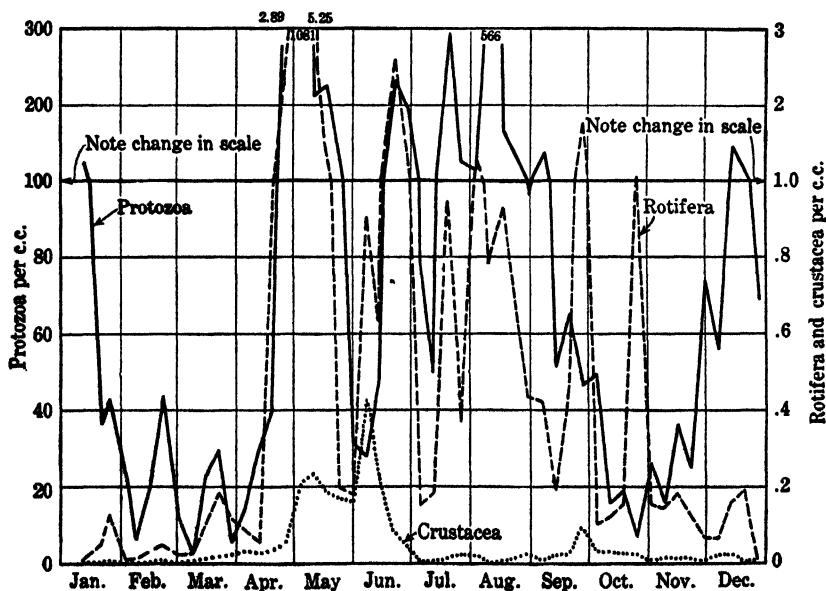


FIG. 87. — Seasonal Distribution of Protozoa, Rotifera, and Crustacea. Illinois River at Havana, 1898. After Kofoid.

in a series of pulses that vary widely in amplitude but relatively slightly in periodicity. In the Illinois River the vernal pulse is particularly marked. In the San Joaquin River the autumnal pulse dominates. While a comparison of catches obtained at corresponding seasons during different years shows extreme variations, it is a matter of observation that the general outline of the annual planktograph of each river remains similar in form in different years. The seasonal distribution of different groups of microscopic organisms in the Illinois River is shown in Figs. 84, 86 and 87.

Dispersion of the Rheoplankton. — The hydrographic instability of the fluviatile environment coupled with the movement of the water commonly produces a fair degree of uniformity of the plankton catches in different regions of a stream except in those areas immediately adjacent to the shores. Even here the ever changing cross-section of a river does not permit the development of as characteristic a littoral flora and fauna as is found along the shores of lakes and reservoirs. Current velocities, too, discourage the differentiation of species.

Transverse Distribution. — The horizontal distribution of the rheoplankton along transverse sections of a river is more marked than that along longitudinal ones of limited extent. This is due to the fact that the environmental changes encountered between mid-stream and shore are relatively significant while longitudinal variations are commonly not found in short stretches. There are of course special situations, such as the entrance of tributary streams, where variations along the river are more pronounced than those across it. In heavily polluted streams the longitudinal variation in the environment produces extreme differences in the plankton catches. This is discussed in the next chapter.

The transverse distribution of total plankton in the Illinois River on August 26, 1896 is shown in Table 72. The river at this time stood at 6.5 feet above low water and had a width at the station of 150 meters. The collections were made with a pump and net, 0.25 cubic meters being strained from bottom to surface. The wind was blowing towards the east shore where *Wolffia* were in great abundance. The west shore was rich in vegetation.

The average departure of the volume of the catch from the mean was 27.2 per cent. The variation in the different species is not recorded but it is safe to say that their distribution, too, would be relatively uniform except in immediate proximity to the shores. Here a dominance of littoral forms would be expected. The average departure obtained in this test is no greater than that observed in Lake St. Clair by Reighard. His collections were made within ten days at 14 different points

on the lake. The average departure from the mean of the volume in cc. per m.³ of water was 28.8 per cent. When these results are expressed as volumes in cc. per sq. m. of surface the average departure from the mean of the Illinois River catches is 38.4 per cent that of the St. Clair collections 31.8 per cent. Elimination from the Illinois test of the shore collections reduces even further the variation in the horizontal distribution of plankton in this stream.

TABLE 72
TRANSVERSE DISTRIBUTION OF PLANKTON
Illinois River, August 26, 1896
After Kofoid

Distance from East Shore, m.	Depth, m.	Temperature, °F.		No. of Organisms per Cubic Meter	Volume of Catch, cc. per m. ³
		Surface	Bottom		
10	1.68	82	77	143,800	2.00
37.5	3.96	78	77	110,000	1.34
75	4.88	77.5	77	95,600	1.34
85	4.88	77.5	77	110,200	1.52
95	4.27	77.5	77	109,600	1.44
105	4.04	77.5	77	93,100	2.36
115	3.18	77.5	77.1	112,500	2.40
125	1.68	77.5	77.5	110,300	1.64
135	1.22	77.75	77.5	109,900	1.04
146	0.56	77.75	77.6	96,200	.60

Longitudinal Distribution. — In testing the longitudinal distribution of channel organisms it is possible to compare either simultaneous catches from two different stations along the river or consecutive catches taken at the same place. In the latter method the interval elapsing between catches results in bringing to the fixed station a new volume of water that could equally well have been sampled at a station higher up. The results of ten consecutive catches from a fixed boat are shown in Table 73.

The average deviation from the mean obtained in this test is only 3.58 per cent. A similar experiment from a floating boat in a short stretch of river yielded an average deviation of 11.2 per cent. When the distance between sampling points is increased the changing environmental factors, river stages, tributaries, and other conditions naturally produce more marked variations. Nineteen collections made in two days and

distributed over 205 miles of the Illinois River from its mouth to Hennepin yielded a deviation of 89 per cent.

TABLE 73
LONGITUDINAL DISTRIBUTION OF PLANKTON
Measured by Ten Consecutive Catches from a Fixed Boat
Illinois River, October 29, 1896
After Kofoid

No.	Volume of Catch, cc. per m. ³	No	Volume of Catch, cc. per m. ³
1	3.20	6	3.20
2	3.24	7	2.94
3	3.04	8	3.20
4	3.40	9	2.94
5	3.06	10	3.10

The longitudinal distribution of littoral organisms is more variable, even as the shore environment is less constant than that of the channel.

Vertical Distribution. — That the vertical distribution of microscopic organisms in rivers is not so significant as in lakes becomes apparent when we remember that rivers are commonly not as deep as lakes and that water movement and absence of stratification operate against the concentration of organisms in the different water strata. Within the limitations thus imposed upon the plankton, however, we can expect a selective distribution of organisms much as in standing water. The uniformity of total plankton catches from various depths of water recorded in Table 72 is in agreement with this reasoning.

Summary of Kofoid's Illinois River Findings. — The five-year study of the Illinois River made by Kofoid during the last years of the past century constitute America's most noteworthy biological contribution to rheology. Briefly summarized Kofoid's findings that are of more general application are as follows:

1. Temperature affects plankton profoundly. Below 45° the plankton content of the Illinois River falls to about 9 per cent of that present at higher temperatures. In backwaters it drops to 29 to 40 per cent.

2. Light affects plankton production. The half year with more illumination and fewer cloudy days produces from 1.6 to 7 times as much plankton as that with less illumination and more cloudy days. Seasons of unusual cloudiness are accompanied by depressions in production.

3. Fluctuations in hydrographic conditions are the most immediately effective factor in the river environment. Rising levels usually produce

a sharp decline in plankton content as barren storm water mixes with or replaces plankton-rich water in the channel and backwaters. Hydrographic stability is beneficial, instability destructive to plankton production.

4. Area and depth, within the limits of observation, show little relation to plankton production.

5. The dispersion of plankton in the Illinois River is about as uniform as that in German lakes and in Lake St. Clair.

6. There is little correlation between the seasonal flux in chemical conditions (as shown by sanitary chemical analyses) and the seasonal course of plankton production. The nitrogenous matters are influenced by the growth of microscopic organisms, especially by the multiplication of diatoms, but the changes are not uniform or proportional.

7. The age of the water is an important factor in the production of plankton in streams. Young streams have but few plankton. Impounded for 10 to 30 days this barren water develops an abundant plankton crop. The rate of run-off and replacement of impounded waters influences plankton production, being greatest where run-off and renewal are least.

8. The channel plankton is subject to great seasonal and annual variations. The catches of individual years differ greatly as a result of hydrographic, climatic and other environmental variations.

9. The course of plankton production in channel and backwaters exhibits a series of recurrent pulses. This cyclical movement is plainly influenced, accelerated or retarded, or its amplitude extended or depressed, by environmental factors, but is not itself traceable to any one, or any combination, of them. The pulses vary from 3 to 5 weeks in duration.

10. The normal regimen of the course of plankton production in the Illinois River and its backwaters does not describe a definite seasonal planktograph. It consists rather of a series of recurrent plankton pulses, the amplitudes of which are largely determined by the fluctuating environmental factors of the unstable fluvial environment. Planktographs of the same locality in different years and of different stations show resemblances only in certain fundamental features, such as winter minimum and vernal pulse. A more stable environment is conducive to greater relative production of plankton.

11. The plankton method can be applied to a stream as legitimately as to a lake.

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CHAPTER XII

SELF-PURIFICATION OF STREAMS

Surface waters constitute by far the largest class of public water supplies in North America. At the same time they are the ultimate recipients of the water-carried wastes from many cities and industries. A number of our water courses serve alternately as water supplies and sewage carriers, and many large lakes are used simultaneously for these purposes. This dual use is made possible by the power inherent in water to purify itself. It is a power intimately connected with the microscopic organisms that develop in water and as such offers one of the most fruitful fields for microscopic investigation.

"Self-purification" is the natural process or combination of natural agencies that tends to render stable and innocuous foreign substances that find their way into water and so to restore the water to its natural condition of purity. The term is most often applied in connection with streams that have been polluted by sewage or industrial wastes, because it is in such waters that the effects of self-purification are best defined and most conspicuous. The forces involved in the natural purification of streams, however, operate in standing water as well as in running water, though in different degrees of magnitude. They act, too, upon soil wash, decaying vegetation and other matters that are discharged into water courses as well as on polluting substances contributed by human habitations and industrial works.

Forces of Self-Purification. — The forces of self-purification are physical, chemical, and biological in nature. They are closely inter-related and mutually dependent. The general manifestations and methods of operation of these forces have been discussed in connection with limnology, rheology and water storage. In order to avoid repetition, therefore, they are merely outlined below.

Physical Forces. — The most important physical forces are gravity, light, and aëration.

Gravity — Removes heavy suspended impurities by sedimentation of individual particles; removes colloidal matters and light suspended matter by settling of aggregated or coagulated masses.

Light — Bleaches color; induces photosynthesis, increasing dissolved oxygen and removing carbon dioxide.

Aëration — Adds oxygen by absorption from the atmosphere; removes carbon dioxide and other gases of decomposition by escape to the atmosphere.

Chemical Forces. — Outstanding among the chemical forces of self-purification are oxidation and reduction. Chemical coagulation is involved to a lesser degree.

Oxidation — Changes suspended and dissolved organic matter to mineral matter, relatively stable organic matter, or gases. Causes the precipitation of dissolved mineral substances such as iron and manganese.

Reduction — Hydrolysis and splitting chemical processes produce liquefaction or gasification of organic matter, and pave the way for stabilization and mineralization by oxidation. Puts mineral matter into solution.

Coagulation — Causes precipitation of dissolved and colloidal substances by chemical reactions induced by natural processes or the addition of trades' waste.

Biological Forces. — The biological forces of natural purification are closely associated with the food habits of the "living reagents" found in polluted waters.

Bacteria — Attack dissolved and suspended organic and mineral substances and convert them by aërobic or anaërobic digestion into end products of simpler chemical structure.

Algæ — Use up carbon dioxide and produce oxygen. Utilize simple inorganic food substances resulting from bacterial activity and also certain nitrogenous organic substances.

Protozoa — Live on organic matter. Many species prey upon bacteria. Chlorophyll-bearing protozoa act similarly to algæ.

Rotifera and crustacea — Consume algæ and prey on protozoa. Saprophytic organisms feed on decaying materials.

Large aquatic plants — Act similarly to algæ; rooted forms utilize food substances contained in bottom deposits.

Large aquatic animals — "Work over" mud deposits. Insect larvæ utilize food substances contained in the water and the sediments. Fish live on plankton and insect larvæ.

ZONES OF POLLUTION AND SELF-PURIFICATION

When sewage or some similar polluting substance is discharged into water a whole train of events is put into action. If the sewage is discharged into a lake in which the currents about the outfall are sluggish

and shift their direction, these events take place in close proximity to each other, change their location and are not well defined. If, on the other hand, the water is in motion, as in a stream, the events do not occur at one place but in a series of zones readily distinguishable below the point of pollution. These zones of self-purification are not fixed but shift and are modified in character with the changing seasons and with varying hydrographic conditions. They are more accentuated in type during the warmer months and during low river stages than in winter or when the stream is in flood. In spite of this variability, the zonal differentiation is sufficiently marked at all times to establish its value in the diagnosis of the natural purification of streams.

When the pollution of streams is slight its effects as well as those of self-purification are suppressed. Hence it is best to hinge the discussion of natural purification on conditions of extreme pollution in which the different events stand forth in greater contrast and are more readily identified.

Zone of Degradation. — When a large volume of sewage or other organic waste material is discharged into a clear stream the first sequence of events partakes of the following nature.

The sewage or waste makes the water turbid and imparts to it a gray color. Sunlight is shut out; part of the suspended matter settles to the bottom and forms sludge deposits that decompose slowly.

At first the organic matter discharged into the water is fresh and is often relished by fish, which may therefore feed in the vicinity of the outfall. Decomposition, as a result of bacterial activity, however, soon begins; the bacterial flora increases greatly and reduces the supply of dissolved oxygen; carbon dioxide increases.

As decomposition becomes established the typical water fungi appear. Suter has described their development as follows:

They form in dense masses, covering the stones of the bottom and clinging to submerged sticks and plants. In still water they may be bulbous in form, like summer clouds; where there is more current the masses are more shaggy or fleecy with jagged edges and points swaying in the current. Fragments are frequently detached by the current and carried downstream into the lower zones. While submerged, these plants have somewhat the appearance of cotton wool, although made up of gelatinous threads. The newly developed forms are frequently white, but the older parts tend towards an olive green or "putty" gray color, turning to a rusty brown in the oldest portions. These fungi feed on and disintegrate waste material and absorb oxygen. They persist until the oxygen saturation is pulled down to about 45 per cent, 4 p.m. at summer temperatures, when they die and in turn support the lower forms of fungo-bacteria.

Mixed with these fungi, or forming growths of similar appearance, are other microscopic organisms, notably colonial ciliates that in bulk pre-

sent a structure quite like that of the fungi. The fungi represented are chiefly *Sphærotilus natans*, the so-called sewage fungus, *Leptomitus*, and *Achlya*. The ciliates forming furry growths are most frequently *Carchesium*, *Epistylis*, and *Vorticella*. Purdy has estimated the number of individuals in one cubic centimeter of ciliate growth at over 141,000.

Green plants are found in this first zone, but photosynthesis and with it growth is reduced by the occlusion of light. The larger and more highly organized plants persist in the upper portion, the smaller and lower forms in the lower one until they too disappear as the oxygen is used up and the lower limit is reached. Littoral forms of green or blue-green algae cover submerged or frequently wetted stones or mud along the margins and trail into the current from their anchorage much like the fungi. In the grayish water which robs them of sunlight they are frequently gray-brown in color rather than green. Among the organisms found in this environment may be mentioned, *Stigeoclonium*, *Oscillatoria* and *Ulothrix*.

When the current is not too rapid much of the suspended matter carried into the stream by the sewage settles out and forms a pollutorial carpet in which new types of life make their appearance. The most common denizens are small reddish worms (tubificidæ) quite similar to the common earthworms though smaller in size. They are found in very large numbers in bottom sludges of high organic content and decrease rapidly as the condition of the bottom sediments improves. The worms are of several varieties; *Tubifex* and *Limnodrilus* are common species.

It is apparent that this zone of recent pollution is a zone of degradation in which life is reduced from a more highly organized to a more primitive plane and the physical and chemical quality of the water is abased. According to Suter who has studied the problem of pollution from the standpoint of fish conservation the lower limit of the zone of degradation is defined approximately by a dissolved oxygen content that has been reduced to 40 per cent of saturation or about 3.5 p.p.m. at summer temperatures.

This first zone is found below all outfalls that carry organic wastes into water. In streams it occupies a relatively short stretch of water which in the presence of slight pollution is reduced to a small patch of stream bed in the neighborhood of the point of discharge. When the dilution of the wastes is great degradation may not be completed, and the water may be returned to its *status quo* without recognizable evidence of having passed through the characteristic cycle of self-purification.

Zone of Active Decomposition. — This second zone occurs only in heavily polluted water in which the dissolved oxygen is nearly or completely eliminated and aerobic organisms give way to anaerobes. If we accept Suter's limits for the first zone, the second begins with an oxygen concentration at its upper end of 40 per cent, passes through complete sepsis and then gradually climbs back at its lower end to its initial oxygen value. The characteristics of the upper and lower portion are very much alike although opposite in trend. The middle portion is truly septic, i.e. completely devoid of dissolved oxygen. In the absence of gross pollution the upper and lower portions of the zone come together and the stream does not become septic. When pollution is light the zone of active decomposition is not found. In heavily polluted rivers, however, this zone extends over many miles.

From a physical standpoint the zone is marked by: grayish color of the water; sticky blackened sludge deposits with offensive odor; evolution of gas bubbles especially during the warmer months; foul sewage and privy odors.

As long as the water contains dissolved oxygen aerobic decomposition takes place. When the dissolved oxygen is exhausted anaerobic decomposition or putrefaction sets in; carbon dioxide increases; reducing and splitting chemical processes predominate; ammonia and nitrites are high, nitrates low or absent.

Bacteria continue to flourish in large numbers, anaerobic forms taking up the work of the aerobic organisms when the middle portion of the zone is reached. Later they make way for aerobic bacteria as the rate of biochemical oxygen demand is overbalanced by the rate of reaeration. Protozoa and other animalcules follow a similar course to the bacteria, aerobic forms being succeeded by septic genera which are later followed again by aerobes. Bacterivorous and other predatory organisms increase greatly in the lower portion of the zone.

At the upper end of the zone the transition to septic conditions is marked by a reduction in fungus life. This at first is plentiful and similar in appearance to that of the preceding zone. The organisms are more thread-like in structure and develop pink, cream or gray tints that may become very dark. In the portion of true sepsis they disappear.

Green plants are absent except at the lower end of the zone where minute chlorophyll-bearers assist in reoxygenating the water.

Tubifex worms disappear in the upper end of the zone and reappear near the lower one. Rat-tail maggots develop in all but the most septic parts. Larvæ of the sewage fly (*Psychoda*) are found. Fish do not penetrate this zone.

Kolkwitz, Marsson, and others do not recognize the zone of degra-

dation as distinct from that of active decomposition. The first being relatively small in extent is grouped with the larger one. The combined zones were characterized by Kolkwitz and Marsson as "polysaprobic" i.e., life in the presence of much decaying matter.

The delimitation and nomenclature of the zones of self-purification suggested by different observers is shown in Fig. 88.

Clean water	Degradation	Active decomposition	Recovery	Cleaner water
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Suggested classification

Clean water	Recent pollution	Septic	Recovery	Cleaner water
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After Suter 1922

Clean water	Septic	Polluted	Contaminated	Cleaner water
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After Forbes and Richardson 1913

Clean water	Septic ?	Pollutional or unusually tolerant	Sub-pollutional or tolerant	Clean water
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After Richardson 1921, 1925

Ohgo-saprobic	Polysaprobic	α -Mesosaprobic	β -Mesosaprobic	Ohgo-saprobic
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After Kolkwitz and Marsson 1911

FIG. 88. — Zones of Self-purification as Designated by Various Workers.

Zone of Recovery. — The zone of recovery is directly opposite in its manifestations to the zone of degradation, and extends over much longer stretches. Kolkwitz and Marsson have called it the "mesosaprobic" zone, signifying life in the presence of a moderate amount of decaying matter. Forbes and Richardson have included approximately the last quarter of the zone of active decomposition and the first half of the zone of recovery in a separate zone which they designate as "polluted." This zone according to their standards is followed by a "contaminated" river stretch which corresponds to the last half of the zone of recovery. The polluted and contaminated zones correspond to the α - and β -mesosaprobic zones of Kolkwitz and Marsson. The term "zone of recovery" was suggested by Suter who assigns 40 per cent saturation with dissolved oxygen to the boundary between the middle and third zones. Purdy has suggested values of 15 to 60 per cent for the polluted zone and 70 to 100 per cent for the contaminated one.

The physical character of the zone of recovery is marked by gradually

clearing water and by bottom deposits that are granular rather than sticky and have no offensive odor. Gas bubbles are absent.

Dissolved oxygen increases to complete saturation after the biochemical oxygen demand has been largely satisfied. Free carbonic acid is reduced. Ammonia is present in moderate amounts; nitrites and nitrates increase. Judged by chemical standards this is a zone of mineralization.

Bacteria decrease in number as the available food stuffs are consumed and as predatory organisms develop. Protozoa, rotifera and crustacea are present.

Fungi develop to a limited extent, blue-green and grass-green algæ make their appearance in this order, and diatoms are numerous towards the lower end. Larger aquatic plants reappear. Sponges and bryozoa are found attached to sticks, plants or pebbles.

Tubifex burrows in the mud deposits of the upper regions of this zone, and certain species of blood worms establish themselves. Mussels, snails and insect larvæ develop in the detritus. The more tolerant fish such as the sucker, stoneroller, creek chub and shiner feed on the bottom.

Zone of Cleaner Water. — Below the three zones of pollution and visible self-purification is found the cleaner water zone in which natural purification processes — physical, chemical and biological — continue their beneficent work. This is the oligosaprobic zone of Kolkwitz and Marsson — i.e. life in the presence of little decaying matter. From the standpoint of the sanitarian streams are not completely purified until their waters are fit for human consumption; for this they must be free from pathogenic organisms, as well as attractive in appearance. The lower limit of the zone of recovery commonly falls short of this requirement, and very long periods of flow are necessary to meet it. Hence the dogma set up for the relatively short streams of the British Isles by British bacteriologists a generation ago, "There are no rivers in England long enough to purify themselves."

The cleaner water zone is characterized largely by the normal flora and fauna of rivers discussed in the preceding chapter. Most ponds or lakes, too, constitute an oligosaprobic environment. Green plants large and small predominate. Microscopic animal forms are scarce consisting chiefly of rotifers and crustaceans. The clean bottom is populated with the larvæ of the stone fly (*Perla*), may fly (*Heptogenia*), etc. Mussels and cray fish may be present. Game fish find conditions favorable for existence.

In natural streams the zones here discussed neither occupy a fixed position in the channel nor are they sharply bounded. The zones

overlap due to variations in temperature and river flow which cause a shifting of the zones. Some times they are extended, some times contracted. Thus the zone of active decomposition is at times completely eliminated; at times it is spread over long river stretches. The cycle of self-purification may be retarded, interrupted or accelerated by the entrance of new waste materials, by tributary waters different in nature from those of the main stream, by dams and rapids or stretches of sluggish water. This shifting of the zones may create conditions of existence that are different in the ooze and the water flowing over it. Thus the bottom sediments may be of mesosaprobic character while the supernatant water has become oligosaprobic due to the dilution of the water by high rainfalls. Changes in the stream bottom indeed usually lag behind those in the water. Each river system operates under conditions peculiar to it alone. Correct interpretation of rheological findings hinges upon a thorough knowledge of these conditions.

PARAMETERS OF SELF-PURIFICATION

The search for a parameter that will measure the progress of self-purification has yielded a number of valuable "yardsticks" that will measure the various changes that take place in the course of the natural purification of streams. Some of these, such as the physical appearance and odor of the water, are but coarse measures of existing conditions; others, such as the dissolved oxygen content of the stream, its biochemical oxygen demand, and its bacteriological character, are capable of giving more refined results. Some of the parameters are of particular interest to the ichthyologist, others to the sanitarian. In this connection it is well to note that much of our knowledge relating to self-purification is derived from investigations into the conservation of fish life and that in the United States some of the most fruitful work in this field has been accomplished by Federal and State Conservation Commissions or similar governmental bodies.

Of the parameters of self-purification at our disposal we shall select for discussion only those that are closely related to the study of sanitary microscopy and that yield significant information on the mechanism of natural purification. This mechanism is activated essentially by biological forces, the operation of which, however, is greatly dependent upon the physical and chemical environment in which the activating organisms have their habitat. The reactions accompanying the operation of this mechanism furthermore are manifested in physical and chemical as well as biological changes. As a result the indicators of self-purification may be of a nature other than biological.

Dissolved Gases. — Enough has been said in preceding chapters about the delicate balance maintained between dissolved gases and aquatic life to show that an examination into their use as parameters of self-purification will be fruitful. The dissolved oxygen content of water in particular is a sensitive, rational and readily applicable yard-stick. Its use for this purpose dates back to the earliest scientific investigations of stream pollution in England and it remains today one of the most informative measures of natural purification. Carbon dioxide determinations, too, reflect the changes resulting from the self-purification of water.

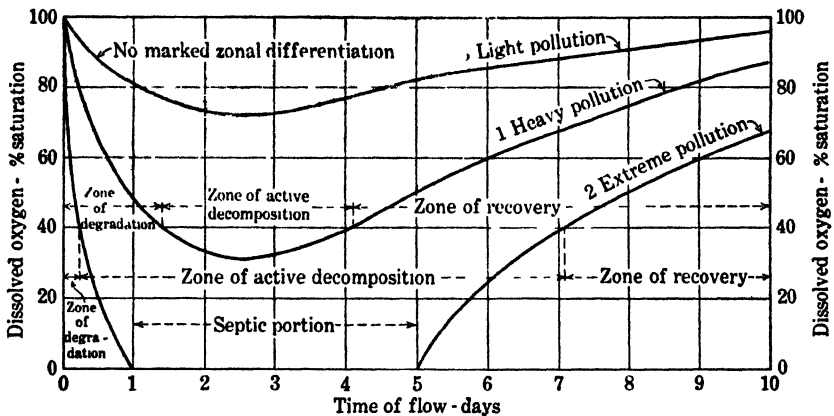


FIG. 89. — Progressive Change in the Dissolved Oxygen Content of a Stream below the Point of Pollution. (Curves are based on the Ohio River Experience below Pittsburgh of the U. S. Public Health Service.)

Oxygen. — When sewage or other putrescible waste matter is discharged into a stream whose waters are saturated with oxygen the resultant changes in the oxygen content of the stream are characterized by curves similar to those shown in Fig. 89. There is an immediate drop in the dissolved oxygen of the stream due to the physical admixture with the oxygen-rich river water of sewage which is usually poor and sometimes entirely deficient in dissolved oxygen. Then follows a gradual depletion in oxygen as a result of bio-activated decomposition of the organic waste materials. At the same time the carbon dioxide increases. The rapidity of the drop in oxygen depends upon the nature and concentration of the wastes undergoing decomposition through the activity of living organisms, as expressed by the rate of biochemical oxygen demand; furthermore upon the rate at which the stream is able to reabsorb oxygen from the atmosphere. This rate of reaeration varies with the nature of the stream, its surface area, depth,

velocity of flow, mixture of the waters etc., and with the relative amount of oxygen in the water. The greater the opportunity of the water to come into contact with the atmosphere the greater the rate of reaëration. Also, the smaller the concentration of oxygen in the waters the greater the rate at which oxygen is reabsorbed from the air.

In a heavily polluted stream the rate of demand for oxygen in the stream at first generally exceeds the rate at which oxygen is supplied from the atmosphere. The resultant oxygen curve (Line 1, Fig. 89) denotes a gradually decreasing oxygen content until a point is reached at which the rate of reaëration just balances that of oxygen demand. This "critical point" is the minimum of the "oxygen sag." It may be close to zero saturation, or lie at a higher value, depending upon the relative amounts of sewage discharged into the water. Beyond this critical point decomposition proceeds at a slower and slower rate as the decomposable matter becomes less; at the same time the oxygen content of the stream gradually increases by reaëration until the water is completely saturated.

As the curve climbs back to higher oxygen values and the stream is able to support the growth of synthetic plankton and large aquatic vegetation, restoration of a condition of saturation is greatly assisted by the photosynthetic activity of these green growths.

In an over-polluted stream the rate of demand is so great that the dissolved oxygen content declines until it is completely exhausted; then septic conditions prevail. Anaërobic decomposition takes place and the river is turned into a dark, putrid stream. As such it continues until the rapidity of decomposition slackens and reaëration gradually builds up the oxygen content as before. In this case the "oxygen sag" is said to "drag bottom." (Line 2, Fig. 89.)

When pollution is relatively light the oxygen curve takes the shape of Line 3, Fig. 89. The locations of the zones of self-purification in relation to the oxygen condition of the streams are indicated for each case. The use of the dissolved oxygen content of a stream as a parameter of self-purification is apparent.

The laws governing the rates of biochemical oxygen demand and reaëration have been formulated by Streeter and Phelps. This makes it possible from a knowledge of certain basic characteristics of the river and the wastes discharged into it to predict the "oxygen sag" that will be obtained. The curves of Fig. 89 are based upon the work of these investigators and the reader is referred to the Ohio River Studies of the United States Public Health Service for a discussion of their methods.

In most streams the oxygen sag is modified by the entrance of tribu-

taries that contribute oxygen-rich water, or by the discharge of putrescible matter into the waters undergoing self-purification. Often, too, the stream is not fully saturated with oxygen when it reaches the

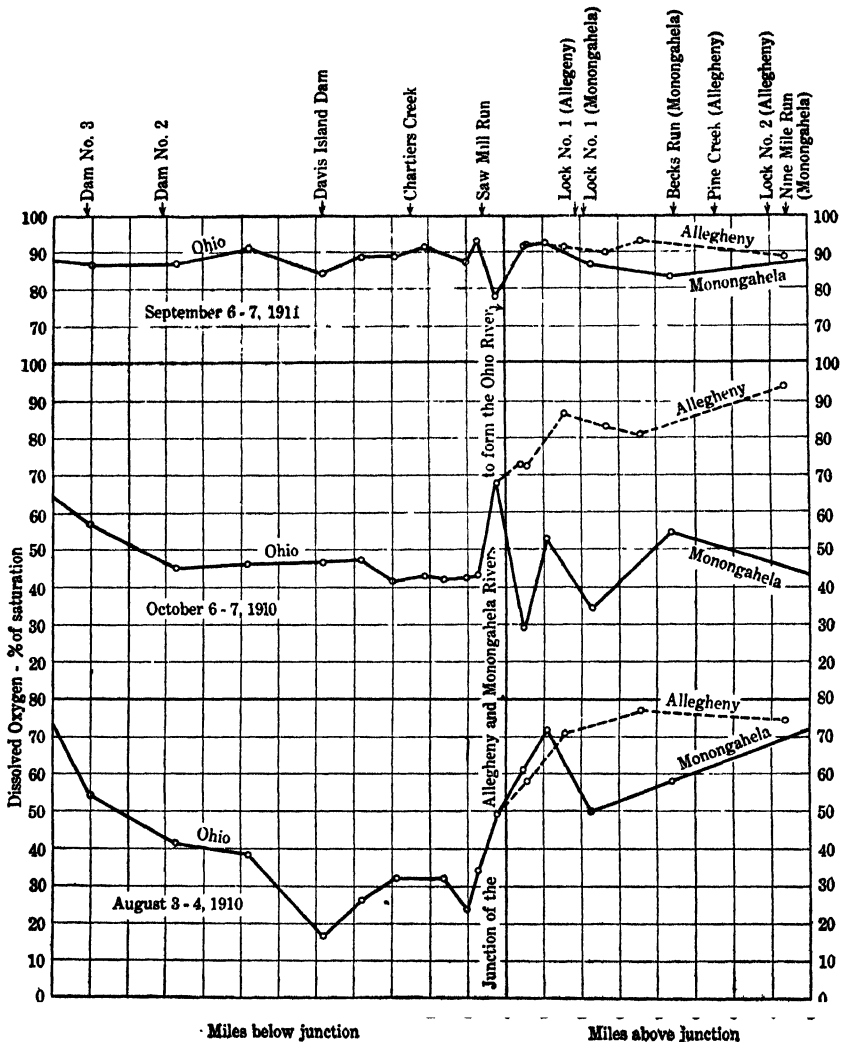


FIG. 90. — Dissolved Oxygen Sag of the Ohio River and its Tributaries at Pittsburgh, Pa., 1910-11. *Courtesy of Allen Hazen.*

point of major pollution. These conditions, too, can be measured and accounted for. The dissolved oxygen sag at Pittsburgh of the Ohio River and its component streams, the Allegheny and Monongahela, was studied by the author in 1910 to 1911. The values obtained are shown

diagrammatically in Fig. 90. These curves indicate the variation of the oxygen depletion under different conditions of temperature and hydrography.

Carbon dioxide. — The factors influencing the CO_2 content of polluted streams are in general the same as those producing the phenomena of deoxygenation and reaeration but are opposite in their action. Thus decomposition, while consuming oxygen, results in the production of carbon dioxide; reaeration, while adding oxygen to the water, removes CO_2 ; and plant growth, while liberating oxygen, uses up CO_2 . Free CO_2 , like dissolved oxygen, is a valuable parameter of self-purification. As yet it has not been as fully investigated as dissolved oxygen and will require further study before it is more generally applied. The variation in CO_2 content of the Genesee River at Rochester is illustrated in Fig. 92.

The important relations of dissolved gases to aquatic life have been pointed out in Chapter VIII. Investigations into the gaseous content of polluted waters, therefore, explain much in the microscopic findings which in turn are an aid to the interpretation of the results obtained.

Mineral Matter. — The inorganic constituents of water are less subject to striking changes during the progress of self-purification than are the gases and the organic content. For the greater part inorganic compounds are not altered in composition by the advent of pollution. The sulphates, bicarbonates and chlorides are often present in considerable amounts before pollution occurs; they may even be appreciably increased by entering wastes, but they are not subject to the oxidizing changes that mark purification. Such salts, therefore, serve a better purpose as measures of defilement than of recovery. It is true that sulphates are sometimes subject to reduction when decomposition is so active as to assume an anaërobic character, and bicarbonates are changed to carbonates when free carbon dioxide is lacking for plant growth. These changes are liable to be too small in magnitude to have great informative value; they have occasional use, as collateral evidence, to support other and more significant observations.

Exception may be taken to some of the foregoing statements in connection with the mineral forms of nitrogen. Due to the successive stages of oxidation through which they pass and to the fact that they are derived from organic impurities, ammonia, nitrite and nitrate nitrogen contribute valuable information regarding the progress of stabilizing forces in polluted waters. Analysts have recognized their value for many years; prior to the development of bacteriology the nitrogen tests, taken together, constituted almost the sole criterion for judging the pollution and purification of water. They will be discussed together under nitrogen and organic matter.

Organic Matter. — It is evident that any test that records definite changes in the character or quantity of organic matter at once becomes a valuable parameter for marking recovery from degrading influences, for it is principally organic material, living and inert, that debases the quality of water. Most of this organic matter, especially that derived from animal sources, contains nitrogen. The latter is fairly easily determined, as are the simpler inorganic forms of nitrogen that result from oxidation. This advantage does not exist with organic carbon; also, carbon passes through fewer intermediate stages during oxidation.

Another method of measuring changes in organic content is that which expresses the amount of oxygen used from a chemical reagent, presumably to oxidize organic matter, during a period of digestion under laboratory conditions. Such a test is the "oxygen consumed" determination employing potassium permanganate.

A third procedure for estimating the amount of decomposable and oxidizable organic material is that which records the amount of oxygen absorbed during a period of incubation, when the active oxidizing agents at work are the bacteria and other organisms common to the sample. This is the "biochemical oxygen demand" test.

Organic Nitrogen. — The acquisition by a body of water of waste organic material, whether from natural sources or from the habitations or activities of man, is accompanied by a rise in the organic nitrogen content, the magnitude of the increase varying with the character and total amount of the waste. The highest nitrogen value occurs at the point of entrance. As time and distance intervene lower values are obtained. Several factors bring this about. Grosser particles settle out, current velocities determining the extent to which this takes place; animal life, macroscopic and microscopic, consumes some nitrogenous material as food; finally the activities of a host of bacteria succeed in splitting, hydrolyzing, and fermenting the complex organic substances with liberation of the nitrogen in the form of ammonia compounds, the first step in the mineralization of nitrogen. In the absence of further pollution there is a steady decrease in organic nitrogen, principally due to the bacterial activity, until the clean water zone is reached when only small amounts remain, most of which are highly stable or in the form of living cells, for the greater part those of the plant kingdom. Time is the determining factor in the observed reduction. The amount of organic nitrogen is, then, a measure of the decomposable material present at any time or place, and so becomes an indicator of the self-purification effected within a given time or distance.

Sewage will contain from 8 to 20 p.p.m., or more, of organic nitrogen and polluted streams below the entrance of sewage from a few tenths to

a few whole parts per million. The test for this form of nitrogen will record considerably less than 0.1 p.p.m., and is sufficiently delicate to indicate the changes that are going on in polluted water. The most rapid reduction occurs where bacterial activity is greatest, i.e. in the zone of active decomposition.

The reduction in organic nitrogen that takes place in relatively short distances of stream flow is illustrated by Table 74. The samples were taken from a small stream in Massachusetts that received the effluent of intermittent sewage sand filters.

TABLE 74
REDUCTION OF ORGANIC NITROGEN IN A POLLUTED STREAM
November, 1926

Station	Distance in Feet from Station 1	Organic Nitrogen in p.p.m.
1	...	0.942
3	1300	1.785
4	2500	1.557
5	4250	0.700
8	7950	0.985

Stations 1 and 3 were both located just below the points of effluent discharge. There was no further pollution from sewage beyond Station 3. The increase in nitrogen at Station 8 was due to the presence of plankton life and to débris from decaying vegetation.

Attempts to use organic nitrogen as a parameter of conditions is sometimes fraught with difficulties. Successive increments of pollution maintain high nitrogen values and mask the purification that has been accomplished. Natural drainage also adds large amounts of organic matter; fluctuations in stream flow modify concentration. Under such circumstances the determination can be of value only when frequent stations are maintained or frequent samples taken. Also, too high dilution of waste reduces the nitrogen to insignificant values. Frost and Streeter concluded from the Ohio River studies of the United States Public Health Service that the nitrogen determinations, as a group, "are not sufficiently precise to measure the effect of the sewage from a city of 500,000 population upon the Ohio River at moderate and high stages. . . . It is only in periods of low discharge in the river, when the effect of comparatively constant sewage flow is proportionately in-

creased, that the effect becomes distinguishable." They further state, "except during the periods of high discharge there is a fairly consistent decrease in organic nitrogen from Pittsburgh to station 461,* above Cincinnati; and an increase, though slight, in passage past Cincinnati, and again in passing Louisville."

Mineral Nitrogen. — In the presence of active decomposition a large amount of ammonia nitrogen, or free ammonia, is formed. This is not an exact measure of the amount of nitrogenous organic matter that has been decomposed, for production of ammonia compounds is not cumulative; they are constantly transformed by oxidizing forms of bacteria into nitrites and nitrates. In the zone of degradation ammonia values increase, reaching their maximum in the upper portion of the zone of active decomposition. At the same time the nitrates reach minimum values and often totally disappear. Nitrites disappear in the presence of anaërobic decomposition; usually they are present in the zone of active decomposition reaching a maximum soon after the ammonia nitrogen. Nitrites, as a rule, constitute only a small portion of the total nitrogen. Thus the rise of ammonia values occurs during the period when organic matter is largest in amount; their fall marks the passing of the most active decomposition and a reduction in organic content. The organic nitrogen and ammonia nitrogen results are sometimes added together and used to represent the unstable, readily decomposed nitrogenous matter plus the nitrogen that has come from such complex material but is not yet converted to stable compounds. The sum of the two is a measure of nitrogen in a state of rapid change.

In the zone of recovery striking changes manifest themselves. Ammonia nitrogen becomes less in amount; it is consumed by plants and oxidized by bacteria. Ammonia production has, of course, not ceased, for organic nitrogen is still present, but production lags behind oxidation to stable forms. Nitrites, too, decline and nitrates exhibit a marked rise. The increase in nitrates may continue into the zone of cleaner water; very often there is a sharp decrease owing to the appearance of plant organisms.

The tests for mineral forms of nitrogen are extremely delicate, and they are excellent parameters of the progress of oxidizing changes. As is the case with organic nitrogen their usefulness is impaired by the advent of fresh pollution, by fluctuating stream conditions and by high dilution.

A typical example of stream conditions with respect to nitrogen content is given in Table 75. The Genesee River at Station 3 was fairly clean, contained practically no sewage or other waste and was super-

* 461 miles below Pittsburgh. ⁴

saturated with oxygen. The station was above a dam where quiet water conditions permitted an abundant plankton growth. The high value for organic nitrogen and the low nitrates at this point were attributable to this growth. Other nitrogen values were low.

TABLE 75
NITROGEN VALUES IN THE GENESEE RIVER AT ROCHESTER, N. Y.
August 13, 1912
(Results expressed in parts per million)

Determination	Station 3 8.5 Mi. Above Mouth of River	Station 2 2 Mi. Above Mouth of River	Station 1 at Mouth of River
Organic Nitrogen.....	1.080	0.838	0.228
Ammonia Nitrogen.....	0.056	1.260	0.780
Nitrite Nitrogen.....	0.002	0.014	0.006
Nitrate Nitrogen.....	0.10	0.00	0.05
Dissolved Oxygen (Per Cent saturation).....	113%	24%	41%

Between Station 3 and Station 2, at a point about 5.5 miles from the mouth of the river, a large trunk sewer discharged into the river. At other points above and below smaller volumes of sewage and industrial waste were discharged. Below Station 2 there was practically no pollution entering.

Station 2 was clearly in the zone of active decomposition, as indicated by the oxygen content. Organic nitrogen, ammonia nitrogen and nitrites were all high, although considerable organic nitrogen was converted to ammonia before reaching this point. Nitrates were absent.

At Station 1, two miles below Station 2, which may be said to represent the upper limits of the zone of recovery, oxygen showed 41 per cent of saturation. Organic nitrogen was markedly lower, and there was a substantial decrease in ammonia and nitrite nitrogen. Nitrates were again in evidence, although held to a low value by plankton growth.

Oxygen Consumed. — This test is a very old one and is designed to measure the amount of oxidizable organic matter present by noting the amount of oxygen consumed in a given period of time, and at a given temperature, from a solution of potassium permanganate. The test is fairly sensitive, but does not register the total oxygen capable of being consumed over a protracted period of time. It has a tendency to indicate carbonaceous rather than nitrogenous material, and so in sewage

and heavily polluted waters gives proportionately lower values than in waters containing little sewage but large amounts of organic matter from natural sources. There are many conditions of the test that require rigorous control in order to produce comparable results from different workers.

Extensive use was made of the oxygen consumed test in the Ohio River Studies of the United States Public Health Service, and its value was closely examined in comparison with total nitrogen as a measure of stream pollution. When applied to a wide set of observations, it was found that the range of variation in oxygen consumed was somewhat narrower than in total nitrogen. "On the whole, oxygen consumed values in the Ohio River are a somewhat less sensitive index of sewage pollution than total nitrogen, and while the results of the two determinations are generally consistent, it appears that the determination of oxygen consumed yields little if any information of value that is not given by nitrogen determinations."

Oxygen consumed finds its best use as a parameter of purification when it is applied to a study of those waters in which are found the typical zones of self-purification. Here the changes in organic matter are large, and the test is delicate enough to measure the stabilizing effects produced by the natural forces that are at work.

Biochemical Oxygen Demand. — In the light of present knowledge the best indication of the status of self-purification, so far as any one test can determine it, is afforded by the results of the "biochemical oxygen demand" test. This calls for incubation of a sample, with or without dilution, at a controlled temperature, usually 20° C., over a definite period of time, from 1 to 20 days, and in the presence of an amount of dissolved oxygen that will provide an excess at the close of the incubation. This excess will be over and above the demand of biochemical reactions resulting from the activity of bacteria or other organisms that are common to the sample. Their principal food is the existing organic matter. The amount of dissolved oxygen present before incubation minus that present at the close of incubation is a measure of the oxidizability of the organic impurities present, or of the demand that will be put upon oxygen resources by natural digestion of organic matter.

The test employs no reagents or forces uncommon to the sample in Nature's laboratory; it is based upon the fact that in the presence of a supply of oxygen, oxidizing bacteria and other organisms, and organic matter capable of oxidation, decomposition will take place with progressive oxidation and stabilization of the organic matter; further, that the rate at which the biochemical oxygen demand is satisfied under

natural conditions is governed by the same law that controls the rate under laboratory conditions. This latter fact has been amply demonstrated by Streeter and Phelps in connection with the phenomena of oxidation and reaeration in the Ohio River studies of the United States Public Health Service. The statement of the law is: "The rate of biochemical oxidation of organic matter is proportional to the remaining concentration of unoxidized substance, measured in terms of oxidizability." Thus, if 10 per cent of the initial oxygen demand is satisfied in 24 hours, 10 per cent of the residual demand will be satisfied in the following 24 hours. The law has been formulated by Streeter and Phelps and enables prediction to be made of the biochemical oxygen demand during different periods and at different temperatures of incubation.

Determination of the oxygen demand does not attempt a direct measurement of any specific organic compounds or classes of compounds, or any portion thereof; it is concerned only with the tendency of any such compounds to be decomposed and oxidized in the presence of bacteria and other organisms and of oxygen. In other words, it deals with organic stabilization processes. It indicates the ability of certain material forces to deal with degrading influences and amplifies the determination of dissolved oxygen, in that it measures the extent of use to which the oxygen will be put in effecting purification and permanent changes.

There is one important influence that the laboratory procedure neglects; this is the reaeration of the water by the oxygen of the air, a subject that has previously been discussed.

For comparative purposes, and for marking the progress of self-purification in streams, laboratory results of oxygen demand tests provide a most useful parameter when there is no great contribution of diluting water or waste between points of sampling. Marked changes in the volume of flow or large additions of fresh pollution tend to mask the extent to which stabilization processes have gone forward. In the Ohio River studies it was found that a much better idea of the changes between stations was obtained if the oxygen demand values were multiplied by the flow of the stream expressed in thousand second-feet. The resulting "quantity units," i.e. "second-foot-thousandths," indicate the amount of oxidizable material corrected to a common basis of dilution, which is carried past given points in a unit of time, the amounts being measured in terms of oxygen demand. The second-foot-thousandth figures may be expressed as grams per second if multiplied by the factor 28.317. An increase in quantity units registers the incoming of new oxidizable organic matter from tributaries or from drain-

age; a decrease is evidence that self-purification is proceeding fast enough to more than take care of additional pollution.

The actual conduct of tests to determine biochemical oxygen demand requires more than ordinary attention to control of conditions and to matters of technique. Sampling must be carried out with care and at points where local conditions do not modify the representative character of the sample. The laboratory procedure must take note of numerous places where errors are liable to be introduced through improper reading or control of temperatures, through aeration of samples, preparation of dilutions, use of unclean apparatus and the like.

If the dilution method is employed, which is commonly the case, it makes use of the modified Winkler method for the determination of dissolved oxygen. Many inconsistencies in final results are attributable to disregard of precautions with this method. Theriault has fully discussed these in his paper, "The Determination of Dissolved Oxygen by the Winkler Method."

When the oxygen demands of different concentrations are used to calculate the demand of the undiluted sample a series of different values often results. Usually the highest concentrations give the lowest values. This seeming inconsistency was investigated by Theriault and Hommon* who showed that the results obtained by incubating different concentrations could be harmonized if the concentrations were increased by a constant. The error was ascribed to a greater oxygen demand of the diluting water when incubated with waste or sewage than when incubated as a blank. The procedure that was proposed to allow for this was the use of diluting water that has been stored until its oxygen demand is less than 0.6 p.p.m. If such water is not used the results from various concentrations may be corrected by the differential method that cancels out the error common to all concentrations. Each concentration is subtracted from the higher concentrations to obtain a corrected concentration factor, and each depletion, without allowance for a water blank depletion, is subtracted from the higher depletions to obtain corrected values. When the oxygen demand of the sample is calculated from these results all concentrations will be in close agreement, provided the tests have been carried out accurately.

Bacteria. — The use of bacteriological parameters of stream pollution and subsequent purification is familiar to sanitarians. Sewage and many other wastes are rich in organic matter and bacteria. Discharged into a stream or other body of water, they supply food for growth and activity, and greatly increase the bacterial flora of the water. This intensifies the biochemical processes and gives rise to complicated re-

* Public Health Bulletin No. 97.

relationships between various groups of bacteria and other low forms of life. Thus there may be examples of true and partial symbiosis, of metabiosis and of antibiosis, activities that are promoted or stifled by the presence of forms not directly responsible for the process.

In heavily polluted streams bacterial activity gradually eliminates free oxygen. In the zone of true sepsis thus created aërobic bacteria largely give place to an enormous variety of anaërobic forms which attack protein substances, cellulose, fats, urea and even mineral salts, such as sulphates and nitrates. The relation of these forms, one to the other, is not well understood, but by a series of symbiotic or metabiotic reactions soluble compounds are formed, such as albumoses, peptones, amino acids, ammonia, methane, and hydrogen sulfide. By successive stages the end products become simpler in chemical structure until finally the environment is unfit for anaërobic activity. Reaëration is then able to overcome the septic conditions, dissolved oxygen is gradually replenished and aërobic forms once more take up the burden of digesting the organic matter; by a sequence of oxidizing reactions nitrogen end-products are carried to nitrites and finally to nitrates, those of sulphur to sulphates, and those of carbon to carbon dioxide, thus completing the cycles of these elements in nature.

There is no sharp division of time as to when all these complicated changes begin and end. There is over-lapping, but the final result is the conversion of complex material to simple, soluble and stable compounds, and the destruction of myriads of bacteria through the elimination of requisite food supply. It must not be forgotten that agents other than bacteria aid in this digestion of organic matter in the water itself. Some of the algæ and plants and many forms of protozoa carry the process forward. In so doing they exhibit an antibiotic relationship to the bacteria, depriving them of food. This antagonistic influence may even take a more active form through the production of end-products that are bactericidal in effect. Also, bacteria are ingested as food by some species of protozoa.

The elimination of organic substances from the water has its counterpart in the sludge and ooze that collect on the bottom. In fact, a greater bulk of solids is digested there than in the water itself. Were it not for this, streams and ponds would soon fill up, just as the surface of the earth would soon become clogged and all life extinguished, if dead plant and animal bodies and their wastes were not subjected to disintegration and oxidation.

If the identification and cultivation of the many species of bacteria that take part in the decomposition of organic waste were less complicated and time-consuming, bacterial parameters of self-purification would

probably excel all other measures. At the present time, however, only the methods of quantitative bacteriology used in routine sanitary water analysis are readily available. These are three in number described in part by Frost and Streeter as follows:

1. "The gelatin count at 20° C. which measures a heterogeneous group of bacteria under conditions of substratum and temperature selected with a view to favoring the growth of organisms which find their optimum environment in nature outside of the animal body, within the usual range of temperature."

2. "The standard agar count at 37° which is a measure of another heterogeneous group, developing under conditions which presumably are more favorable to the multiplication of bacteria having their natural habitat and optimum environment in the bodies of warm blooded animals and adapted to body temperatures" This test overlaps to a certain extent with the 20° count, as many organisms are able to develop at both temperatures and on both culture media.

3. The test for *Bact. coli* of fecal origin which is the standard bacterial indicator organism of sewage pollution and which presumably does not multiply outside of the human and animal body. The fate of this organism characterizes the self-purification of the water from parasitic bacteria such as the pathogenic organisms of typhoid, dysentery and cholera which originate in the human body. From the standpoint of the hygienist the fate of pathogens is the most important consideration in self-purification.

None of these common tests reflect the changes in the actual total bacterial flora of the stream. They merely show the number of organisms that will develop under certain conditions of cultivation. The large group of anaërobic forms is not isolated. As a result the zonal differentiation so well indicated by the oxygen sag for example is not apparent. Following the maximum development or dispersion of bacteria a short distance below the outfall all three tests show but a gradual diminution of the organisms cultivated, the line of reduction approaching the die-away curve, or curve of disinfection or organic death. This curve plots approximately as a straight line on semi-logarithmic paper. (See Fig. 94.)

The use of these three tests as bacterial parameters of self-purification, although of greatest importance to the hygienist, is not equally valuable to the microscopist. He is more directly interested in measures of the changing conditions of existence that so largely determine the growth of microscopic organisms and are reflected by it. Purdy has attempted to define the bacterial content of the zones adopted by Forbes and Richardson as follows:

Septic or saprobic zone: Very high 20° count. *Bact. coli* in 1/10,000 cc. or less.

Polluted zone: Bacterial count high. Bact. coli present in 1/10 to 1/1000 cc. as a rule.

Contaminated zone: Count at 20° moderate, ranging from 100 to 5000 per cc. Bact. coli in 1 cc. and sometimes in 1/10 cc.

Cleaner water zone: Total count at 20° may range from 100 to 1000 per cc. Bact. coli usually present in 10 cc., but not in 1 cc.

Kolkwitz places the following bacteria in the polysaprobic zone: *Streptococcus margaritaceus*, *Micrococcus ureæ*, *Sarcina paludosa*, *Proteus vulgaris*, *Escherichia coli* (Bact. coli), *Bacillus subtilis*, *Pseudomonas fluorescens*, *Spirillum volutans*, *Spirillum undula*. The larger of these organisms are visible with the magnifications commonly used in plankton microscopy. They are useful indicators of sewage pollution. Some of them are confined to the zone of active decomposition, others are transported into cleaner water.

The rôle of bacteria in preparing food substances necessary for the growth of microscopic organisms and the consumption of bacteria by predatory microorganisms links closely the study of all microscopic life in waters. The variation of bacteria and plankton in a polluted stream is shown in Table 76.

TABLE 76
BACTERIA AND PLANKTON
Illinois River, October, 1921, to August, 1922
After Purdy

Station (Miles Above Mouth)	Volume of Flow (1000 c.f.s.)	Time of Flow from Station 286 (Hours)	Plankton (Cubic Standard Units per cc.)			Bacteria on Gelatin (Number per cc.)
			Pollutional	Cleaner	Total*	
292	8 69		107 51%	105 49%	687	1,913,100
288	8 69		154 49	158 51	673	1,148,780
286	9 21	0	152 60	103 40	637	1,106,500
263	15 46	12	276 65	145 35	881	681,100
277	17 73	36	179 58	132 42	722	219,050
(LaSalle)						
196†	21.14	80	142 45	172 55	932	69,620
179	19.97	135	123 36	221 64	793	38,230
166	20.2	178	88 20	360 80	923	15,015
122	21.6	222	111 20	446 80	1245	25,192
(Havana)						
25	32.4	291	39 16	213 84	1072	13,486

* This total includes "indifferent" organisms, and also those of unknown sanitary significance. † For six months only.

Plankton. — The fact that the plankton are sensitive to changes in their environment makes them good indicators of pollution and self-purification. Larger in size and more readily identifiable than bacteria they offer an attractive means of gaging conditions of existence and with it the sanitary status of streams. Before sanitarians can most effectively put them to this use, however, much work must be done to continue and strengthen their classification as “living reagents” that behave in known ways toward sewage or similar pollution.

According to Purdy the food requirements of the plankton align themselves in general as follows: As a food consumer the plankton is second only to the bacteria. Some of them, notably the ciliates and flagellates, are able to utilize the complex organic substances found in sewage-polluted water. Other organisms, such as the crustacea, are regarded as scavengers because they feed on partly decayed fragments of various plant and animal forms. Various organisms, such as certain rotifers, are, in part at least, herbivorous, finding much of their food in the minute phytoplankton. The latter are synthetic organisms and with the aid of sunlight elaborate their food from simple inorganic substances; at the same time they are great oxygen producers. Many of the plankton are predatory in their food habits; certain protozoa feed on bacteria and are in turn ingested by certain crustacea. As in the case of bacteria there are plankton, — flagellates, ciliates, and others — that are able to exist in the absence of free oxygen. They are called anaerobic or septic forms.

While the plankton, therefore, is a product of its environment it also leaves the imprint of its activity on its surroundings and is a potent factor in the self-purification of water.

The status of different plankton organisms as parameters of self-purification is defined in Chapter XXXII where an ecological classification of the various species will be found. The organisms included in this compilation were selected from the available literature on the subject. They probably represent but a portion of the life actually encountered under different conditions of pollution and natural purification, and it is hoped that the coming years will not only add to their number, but also fix more closely, than is possible at present, their relative position and activity in water undergoing self-purification.

In general the zoöplankton rule the grossly polluted river stretches while phytoplankton predominate in cleaner water. This is to be expected when the food habits of these two groups are considered. The former are more largely analytic the latter synthetic. In this connection a comparison of the plankton catches in the San Joaquin

River and the Stockton Channel at Stockton, California, is of interest. The collections from the river represent natural river conditions, whereas the channel catches are typical of organisms in polluted water.

TABLE 77
MAJOR CONSTITUENT GROUPS OF THE PLANKTON IN CLEAN AND POLLUTED
STREAMS
Annual average of semi-weekly and weekly catches
After Allen

	San Joaquin River (Clean)		Stockton Channel (Polluted)		Ratio of Polluted to Clean Water Forms
	No. of Forms	Individuals per m. ³	No. of Forms	Individuals per m. ³	
Schizomycetes.....	1	19,905	2	16,310	.82
Algae:					
Cyanophyceæ.....	18	1,292,680	15	1,603,414	1.24
Chlorophyceæ.....	13	1,499,479	15	1,811,722	1.21
Diatomaceæ.....	59	39,478,317	47	22,609,062	.57
Conjugatæ.....	9	331,687	6	121,653	.37
Total Phytoplankton.....	97	42,622,068	85	26,162,100	.61
Protozoa — total.....	53	3,494,065	75	5,194,416	1.49
Mastigophora.....	20	2,629,112	25	3,708,569	1.41
Rhizopoda.....	9	150,817	13	167,328	1.11
Heliozoa.....	5	257,232	5	370,995	1.05
Ciliata.....	16	447,909	29	946,762	2.12
Suctoria.....	3	8,995	3	762	.08
Rotifera — total.....	43	883,510	55	4,842,320	5.48
Rhizota.....	3	37,181	2	1,509	.04
Bdelloida.....	6	47,105	6	349,312	7.42
Ploima.....	34	799,224	47	4,491,499	5.26
Entomostraca — total.....	7	22,206	7	560,167	25.4
Cladocera.....	4	7,384	3	3,855	.52
Copepoda.....	3	14,822	4	556,312	37.5
Miscellaneous.....	7	10,449	4	369
Total Zoöplankton.....	110	4,410,230	141	10,597,272	2.40
Total Plankton enumerated.....	207	47,032,298	226	36,759,372	.78
Synthetic (chlorophyll-bearing).....	..	45,231,275	..	29,854,420	.66
Analytic (non-chlorophyll-bearing).....	..	1,801,023	..	6,904,952	3.83

The ratio of phytoplankton to zoöplankton in the clean water of the San Joaquin River is 9.7, in the polluted water of the Stockton Channel

2.5. Similarly, the ratio of synthetic to analytic organisms is 25.1 for the river and 4.3 for the channel.

At Stockton the plankton yield of the clean water is greater than that of the polluted water. This is probably due to the fact that the Stockton channel constitutes a zone of recent pollution in which life is being degraded. Further down-stream an increase in plankton life would normally be expected as a result of the large amounts of food stuff that become available. Thus Forbes and Richardson in their studies of the Illinois River following the opening of the Chicago Drainage Canal noted a significant increase in the plankton content at Havana, 184 miles below the entrance of the canal. Comparing their results with those obtained by Kofoid prior to the opening of the canal they state:

No change has recently occurred in the Illinois River system, or in the basin of the Illinois, to account for the increased productivity of its water except the opening of the sanitary canal connecting the Illinois and Chicago rivers at the beginning of 1900. The effects of this occurrence on the plant and animal products of the stream may conceivably have been produced in one or more of these three principal methods: (a) by a mere increase of the waters themselves, which, in so sluggish a stream as the Illinois, with bottom lands so extensive and so widely overflowed by so small a rise of the river levels, will take effect mainly in great expansions of shallow water, long continued or permanently maintained, with muddy bottoms and more or less weedy shores — situations quite capable of producing a relatively enormous plankton as well as an abundant supply of shore and bottom animals and plants; (b) by the addition of increased quantities of organic matter to the contents of the stream in the form of a larger inflow of sewage from Chicago and its suburbs, in condition to increase the plankton by increasing the supply of food available to the minute organisms which compose it; and (c) by the addition to the plankton of the river, of that of Lake Michigan brought down in the waters of the canal.

The efficiency of the first of these conditions is undoubted and that of the second is, generally speaking, quite possible. The importance of an abundance of organic matter in the water as a means of producing a rich plankton is, in fact, so well known that growers of pond fishes in Europe deliberately manure their ponds to increase the supply of food for their fish; and there is considerable evidence, also, that the plankton of the Elbe is largely increased by the sewage of Hamburg and Altona poured directly into that stream.

Table 78 illustrates the increase and seasonal distribution of the plankton in the Illinois River before and after opening the Chicago Drainage Canal.

The general average of monthly averages for the year 1909 to 1910, it is seen, was 5.07 cc. per m.³, the averages for the earlier years being, respectively, 1.39, 2.10, and 3. A comparison of the seasonal plankton growths as well as the hydrographic conditions for the different years shows the closest resemblance between the years 1897 to 1898 and 1909 to 1910. The ratio of 5.07 to 3.00 should, therefore, be taken as best representing the increase due to the opening of the canal.

TABLE 78

ILLINOIS RIVER PLANKTON AT HAVANA, ILL., BEFORE AND AFTER THE OPENING
OF THE CHICAGO DRAINAGE CANAL. 1895 TO 1898 AND 1909 TO 1910

CC. of Plankton per m.³ of Water

After Forbes and Richardson

Month	Before Opening of Chicago Drainage Canal			After Opening
	'95-'96	'96-'97	'97-'98	'09-'10
September.....	1.52	.38	6.33	.10
October.....	.57	1.10	5.94	2.58
November.....	3.02	.02	1.	1.38
December.....	1.19	.76	.56	.38
January.....	.0145	.01
February.....	.01	.04	.27	.21
March.....	.07	.38	.33	2.18
April.....	5.69	5.11	4.40	29.60
May.....	1.30	5.62	11.30	12.27
June.....	.71	.27	3.96	11.89
July.....	1.44	4.78	.58	.23
August.....	1.17	3.65	.91	.06
Averages.....	1.39	2.10	3.	5.07

The variation of pollutional and cleaner plankton species along the course of the Illinois River in 1921 to 1922 has been shown in Table 76. It is indicative of the self-purification accomplished and is closely correlated with the bacterial counts. The ratio of pollutional to cleaner water forms varies from 1.90 at station 263 to 0.18 at station 25, i.e. in 238 miles.

In general, sewage pollution results in depressing the plankton yield of a stream in the upper zones of self-purification and in increasing the catch in the lower ones. Toxic industrial wastes may destroy or inhibit life over long river stretches until the wastes are sufficiently diluted by incoming cleaner waters or neutralized by other wastes to permit the development of a plankton.

Bottom Sediments and Organisms. — Sedimentation has been mentioned as an important factor in the self-purification of water. By it silt and organic débris are removed from the water and settle to the bottom, the silt to remain inert but the organic matter to undergo decomposition. As far as organic matter is concerned, however, sedimentation merely shifts the scene of purification from the water

to the stream bottom. Here worms, young mollusks, and insect larvæ and pupæ are found burrowing and delving in the mud deposits which also harbor vast numbers of bacteria and protozoa.

The character of the bottom sediments found in any stretch of a stream is a reflection of the cumulative variations that have taken place in the supernatant water while the sediments were being deposited. Bottom sludges, therefore, are in some respects more significant of the average condition of a stream than are its flowing waters.

The examination of sludge deposits is commonly limited to certain physical tests, such as color, odor and consistency, and to the enumeration of organisms that make the sludge their habitat. A method of reporting the results of examination is illustrated in Table 79.

TABLE 79
PHYSICAL AND BIOLOGICAL STATUS OF BOTTOM SEDIMENTS, OHIO RIVER AT
CINCINNATI AND LOUISVILLE
After Purdy

Station	No. of Samples	Time Included	Color		Odor	
			Dark to Black	Light or Yellowish	Offensive or Stagnant	Slight, not Offensive
475	47	July 1914 to Dec. 1915	39	8	42	5
598	60	"	25	35	13	47

Station	No. of Samples	Time Included	Consistency		Pollutional Organisms		Summary, Average Number	
			Soft, Sticky, or Slimy	Somewhat Soft, Granular or Tough	Abundant	Moderate Number or Few	Showing Strong Pollution	Showing Moderate and Light Pollution
475	47	July 1914 to Dec. 1915	37	10	39	8	39	8
598	60	"	18	42	15	45	18	42

The food habits of a number of larger bottom organisms or colonizing forms of microscopic fungi are such that they can be used as living indicators of natural purification. Suter has classified them as follows

from the standpoint of fish conservation. For convenience the zones of self-purification occupied by them are bracketed in this list.

Water molds and scums, particularly if of colors other than green, indicate decreasing oxygen — conditions are not favorable and may be worse downstream. (Zone of degradation.) Tubifex marks approximately the limit of fish life. (Lower limit of zone of degradation and upper limit of zone of recovery.) Rat-tail maggots,* if abundant over the whole bed of the stream are an almost certain indication of prohibited pollution. (Zone of active decomposition.) Blood worms† indicate recovery and conversion of wastes into fish food. (Zone of recovery.) Green plants, mosses, silks and nets usually indicate good and improving conditions. (Zone of cleaner water.)

The use of these larger aquatic organisms as indicators of pollution is facilitated by the fact that they can be recognized without the aid of a microscope and that changing hydrographic conditions do not shift their location in the stream as frequently as that of the smaller organisms found in the water itself. A few of the larger "type organisms" found on the bottom of polluted streams are shown on Plate C.

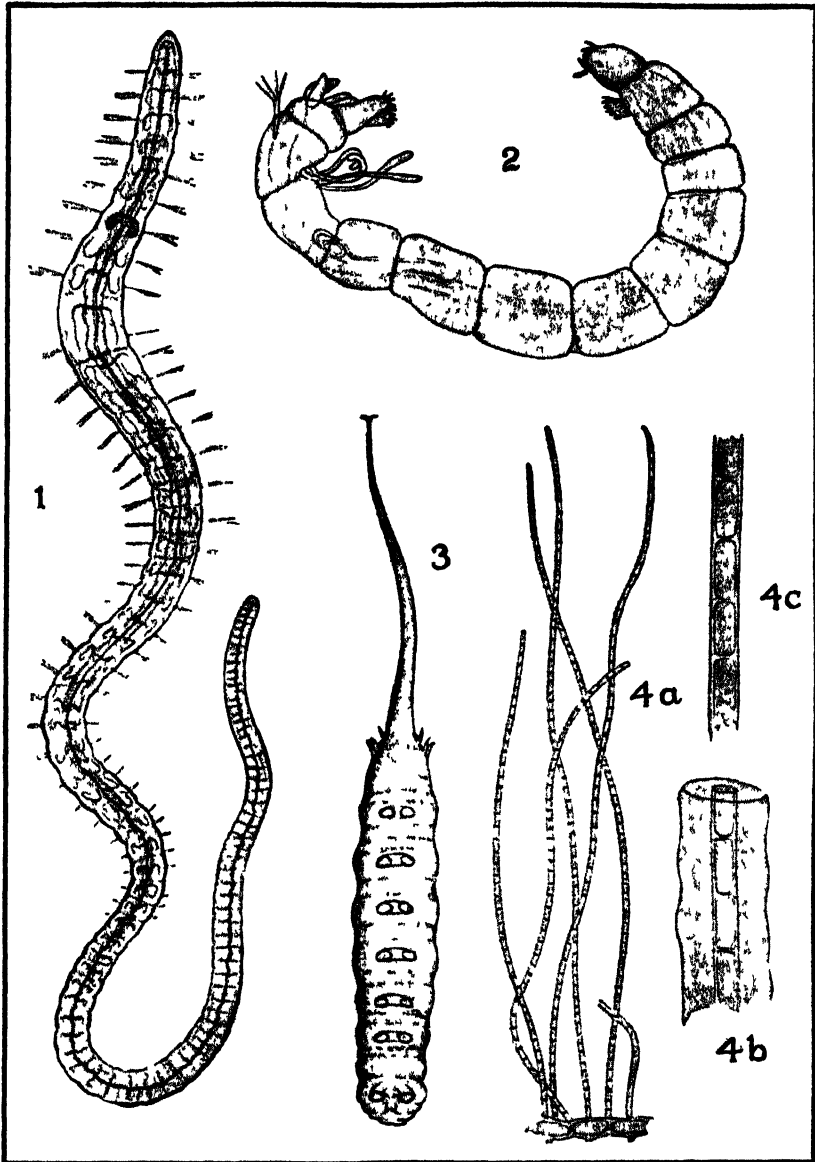
Some species of larger plants and animals are of little value as indicators of pollution because they are found both in clean and polluted water. They are known as indifferent forms and include in their number certain snails, the larvæ of the black fly, and water boatmen.

An extensive classification of the macroscopic indicators of self-purification analogous to that of the plankton is included in Chapter XXXII. The marked reaction of some of the larger aquatic organisms to pollution and the importance of certain members of this group to the self-purification of streams are exemplified by the following two studies.

Richardson's Studies of the Bottom Fauna of the Illinois River. — Richardson has studied the effect of increasing pollution on the bottom organisms of the Illinois River. In 1915 the upper limit of wholesome conditions in the bottom muds was found to be at Chillicothe. During the five years elapsing between 1915 and 1920 Richardson determined from examination of bottom deposits that the added river mileage that was more or less seriously damaged was not less than eighty miles and that the average southward encroachment of pollution thus amounted to

* *Eristalis tenax*.

† Richardson has shown that certain species of blood worms (Midge larvæ, chironomidae) prefer a strongly pollutional habitat while other species are tolerant or indifferent and still others are confined to cleaner water. The use of these organisms as parameters of self-purification, therefore, hinges upon the identification of these various species, a difficult task even for the trained biologist. Kolkwitz and Marsson have made similar reports, and assign the red species to the zones of greater decay, the yellow ones to the oligosaprobic regions.



E. Purkyně del.

PLATE C.

"Type Organisms" found on the Bottom of Polluted Streams.

1. *Tubifex tubifex* $\times 5$;
2. *Chironomus plumosus* $\times 10$;
3. *Eristalis tenax* $\times 5$;
4. *Sphaerotilus natans*:
 - a. Tuft of filaments $\times 400$;
 - b. Cells in both sheath and slimy envelope $\times 2000$;
 - c. Cells in sheath only $\times 2000$.

sixteen miles per year. Summarizing the changes that occurred in this time in the eighteen mile stretch about 150 miles below Lake Michigan he notes: “ (1) disappearance of most species and genera and of several families of small mollusca, along with important average decrease in numbers of the more tolerant forms still surviving; (2) enormous increase in larval midges (chironomidæ), with invasion of several more or less polluttional species, and similar or even greater increase in sludge worms (tubificidæ); and (3) disappearance throughout Peoria Lake, except immediately along shore or in the short, half-mile stretch of swifter water in Peoria Narrows, of all other insects (ephemeridæ, odonata, phryganeidæ, corixidæ, etc.), as well as of planarians and leeches, amphipoda, isopoda, sponges and bryozoa.”

The change in the catches obtained in Upper Peoria Lake is shown in the following table.

TABLE 80
CHANGES IN THE BOTTOM FAUNA OF THE ILLINOIS RIVER, UPPER PEORIA LAKE,
CHILLICOTHE TO SPRING BAY, 1915 AND 1920
Average Valuations, Pounds per Acre
After Richardson

Organisms	Channel		4-7 Ft. Zone		1-3 Ft. Zone	
	1915	1920	1915	1920	1915	1920
<i>Snails</i>						
Viviparidæ and Pleuroceridæ . . .	317	0	87.5	0	50	72
Sphaeriidæ.....	76	56	87.9	12	6.2	12
Amnicolidæ, Valvatidæ, etc.....	0.8	0	0.1	0	8.5	0
<i>Blood Worms</i>						
Chironomidæ.....	0.2	182	4.2	128.6	1.8	0
<i>Sludge Worms</i>						
Oligochæta..... (Principally Tubificidæ)	0.3	173	0.9	40	1.3	8
<i>Other insects, worms, crustacea, etc...</i>	1.9	0	3.9	0	7.7	0
Total.....	396.2	411	184.5	180.6	75.5	92

At Havana, about 200 miles below Lake Michigan, a similar shifting of the bottom organisms was noted. Here, however, the snails had

decreased while the worms had not yet established themselves. This is shown in Table 81.

TABLE 81
CHANGES IN THE BOTTOM FAUNA OF THE ILLINOIS RIVER, LIVERPOOL TO
HAVANA, 1915 AND 1920
Average Valuations, Pounds per Acre
After Richardson

Organisms	Channel		4-7 Ft. Zone	
	1915	1920	1915	1920
<i>Snails</i>				
Viviparidæ and Pleuroceridæ.....	5156	50	319	26
Sphaeriidæ.....	0	17	1777	15
Amnicolidæ, Valvatidæ, etc.....	0.1	0	0.2	0
<i>Blood Worms</i>				
Chironomidæ.....	0	164	0	45
<i>Sludge Worms</i>				
Oligochæta.....	0	3	0	0.1
<i>Other insects, worms, crustacea, etc...</i>	24	6	26	1
Total.....	5180	240	2122	87

During recent years Richardson has continued his studies of the Illinois River and has noted the continued southward movement of pollution.

Purdy's Studies of Limnodrilus. — The tremendous amount of work accomplished by the bottom organisms has been investigated particularly by Purdy, who writes as follows about *Limnodrilus*, one of the sludge worms:

My own personal observation and study on three large polluted rivers (Potomac, Ohio, and Illinois) indicate that these worms are found in very large numbers in bottom sludges of high organic content as shown chiefly by the very repulsive odor. In sediments having little odor, further downstream for instance, these worms occur in rapidly decreasing numbers. Apparently they prefer an environment of heavy pollution. This is strikingly shown in the Ohio River, these worms being very numerous just below Pittsburgh, and again coming into some prominence below Cincinnati, but are practically absent in the lower part of the river near Paducah. The situation is similar in the Potomac, these worms occurring in large numbers just below the sewer outfall, but decreasing until there is only an occasional one forty or

fifty miles below. The Illinois River tells essentially the same story. From an average of 2200 per liter of mud collected near Chillicothe they decrease to 46 per liter at Havana, 57 miles below, and to 12 per liter at Kampsville, 150 miles below. (These Illinois figures are averages from about 30 mud samples from each location, collected during a period of fourteen months.)



FIG. 91. — Work Done by the Sludge Worm *Limnodrilus*. Fecal Castings Deposited Overnight by about 20 Worms. After Purdy.

These worms are interesting and possibly very important biological workmen. They burrow in various directions in the upper layers of bottom sediment, their posterior end meantime protruding a half inch, more or less, above the mud surface, and waving to and fro. They ingest the subsurface mud, and an indication of the amount of work done is furnished by the total quantity of fecal pellets, these being discharged at intervals of about four minutes. The total length of these pellets thus

evacuated during twenty-four hours by one worm is about 69 inches, and this is approximately 45 times the length of the worm itself. There are few parallels, if any, of such *excretory efficiency extraordinary*, relatively speaking, in the known animal world. (See Fig. 91.)

The possible and probable importance of this worm in the economy of stream purification is three-fold, first, its *ability* to work in a nauseous environment which seems to repel most other forms of animal life. They are pioneers, so to speak, in a virgin soil. Second, the *large amount* of work done by them as a result of their great numbers and their continuous activity; and third the *kind* of work done and the net results in terms of stream purification. The *subsurface* mud is elevated and dropped into loose piles of pellets on the surface. Each pellet has a relatively large surface area, and is subject, to a greater degree, to any agencies of purification in the surrounding water than would be the case if this particular bit of mud were still lying undisturbed an inch or so beneath the general surface. A tentative trial indicated that the mud that still remains undisturbed under the surface has a twenty-four-hour oxygen demand more than twice as great as that of these fecal pellets.

Indicator Organisms. — Satisfactory use of aquatic organisms as indicators of pollution and self-purification of water is determined upon the development of an ecological system of classification that will show the normal habitat of all organisms and indicate as well their sensitivity to varying conditions of pollution. Such a system has not yet been established. Enough work, however, has been done to permit the compilation of a fairly extensive list of the pollutional characteristics of organisms frequently encountered in waters undergoing natural purification. Some of these organisms are extremely sensitive to changes in their environment and are, therefore, suited for use as "indicator organisms" or "type organisms" of pollution and self-purification. An extensive ecological classification will be found in Chapter XXXII. In this list definitions of the various saprobic regions in which the different organisms occur are those of Kolkwitz and Marsson. These regions have been discussed above. Their characteristics, as described by Kolkwitz and Marsson, are restated in Chapter XXXII.

In making use of ecological classifications little reliance can be placed upon the occurrence of a single indicator organism. In this respect biological analysis is no different from chemical analysis. A multiplicity of indications is required for a reasonable interpretation of the pollutional status of the water. Instead of considering the different indicator organisms separately, therefore, they should be examined as biological associations that by mutual admission and exclusion of evidence indicate the correct conclusions to be drawn.

The finding of polysaprobic organisms, for example, presents important evidence of the pollutional condition of a stream, but the absence of cleaner water forms is likewise a valuable measure of existing conditions. This latter evidence though negative in character must not be overlooked.

The criteria to be applied to biological associations found in lakes and streams are outlined in Table 82. The characteristics of pollutional associations here given should be of assistance in the interpretation of biological findings. The evidence presented by biological associations

TABLE 82
CRITERIA OF BIOLOGICAL ASSOCIATIONS
After Hentschel

Characteristics	Degree of Pollution				
	Very great	Great	Moderate	Small	Nil
1 Density (Number of individuals)	often great	very great	great	moderate	often great
2. Variety of genera	very small	small	moderate	great	very great
3 Dominance of different genera	very great	very great	great	not marked	generally small
4. Food producers (Synthetic)	almost absent	insignificant	insignificant	many	dominant
5 Food consumers (Analytic)	present alone or very dominant	very dominant	commonly dominant	not dominant	in the minority
6 Bacteria eaters	infrequent to massed	massed	frequent	moderate	infrequent
7. Plant eaters	generally absent	infrequent	not infrequent	frequent	frequent
8. Animal eaters	infrequent	not infrequent	frequent	frequent	very frequent
9. Scavengers	very frequent	massed	very frequent	frequent	few
10 Oxygen requirements of the organisms	slight	very small	small	moderate	great
11. Multiplication and destruction	often very great	very great	great	generally moderate	varying
12. Changes in association	frequently catastrophic	frequently catastrophic	variable	moderate	slow and uniform

and pointing towards "extreme pollution," for example, are as follows: Apart from the fact that indicator organisms must be found their mode of occurrence must partake of a particular nature. They must occur in masses; the variety of organisms must be small; only a few genera

must predominate; synthetic organisms must be almost absent while analytic organisms are preponderant; plant and animal eaters must be infrequent, and scavengers must occur in large numbers; the oxygen requirements of the organisms found must be slight; growth and death must be intense and changes of association must occur with catastrophic violence.

EXAMPLES OF SELF-PURIFICATION

The character and potency of the forces of self-purification of water are best understood by considering some of the outstanding investigations that have been made from time to time in connection with problems of water supply, sewage disposal, disposal of industrial wastes, and fish conservation. It is impossible to treat these investigations adequately within the scope of this chapter. Although summarized below the reader is urged to consult the original sources for an adequate exposition of the factors involved in each case.

Genesee River. — In the summer of 1912 the author studied the pollution of the Genesee River and the shores of Lake Ontario at Rochester, New York. At that time all of the sewage of Rochester was discharged untreated into the Genesee River a short distance above its mouth. The main sewer outfall was $5\frac{3}{4}$ miles from Lake Ontario and self-purification was greatly hastened by the effect of the pure waters of the lake which at times penetrate far into the river. The conditions encountered, therefore, are typical in part of flowing streams, in part of large lakes.

Physical and Chemical Conditions. — The physical and chemical condition of the river on July 15, 1912, is shown in Table 83.

Attention is called to the dissolved oxygen sag induced by the pollution of the river. A similar sag obtaining on July 24 is shown in Fig. 92 together with the variations in carbon dioxide. The recovery of the river as it discharges into Lake Ontario is apparent. Were it not for the admixture of the lake water with the river water the recovery would probably be much slower and extend over a greater distance. The value of nitrogen determinations as parameters of self-purification is well shown. The paucity of information derived from the common physical tests is also evident.

Biological Conditions. — The biological conditions obtaining in this polluted but quickly recovering river are also illustrated in Fig. 92. Quoting from the report made by the author to the city authorities:

For convenience the results of the microscopical examinations of the river water have been summarized according to their natural classification, namely, (1) bacteria, (2) algae, (3) protozoa, (4) rotifera, and (5) crustacea.

Sedimentation is only one method by which the bacteria in the river are removed

TABLE 83
PHYSICAL AND CHEMICAL CONDITION OF THE GENESEE RIVER
July 15, 1912
(Results are parts per million unless otherwise stated)

Station, Miles from Lake Ontario	Depth, Feet	Time	Temperature, °F.	Turbidity	Color	Odor	Dissolved Oxygen		Cl	Total N	Free Ammonia	Nitrites	Nitrates	Alkalinity	Putrescibility, 4 days	Bacteria	Bacterium Coli Present in					
							p.p.m.	% Sat- uration									.0001 cc.	.01 cc.	1 cc.	1. cc.	10. cc.	
.25	0	2:00 P.M.	77.5	8	22	3M	3.32	30.6	42.5	2.800	.700	.040	.35	121.0	Stable	9,000	0	+	—	—	—	
...	18	...	70.0	5	11	3M	9.6	103.0	20.5	2.280	.220	.020	.35	98.0	"	
.5	0	1:45	77.5	8	24	3v, 2M	3.15	31.8	62.5	
...	20	...	70.0	7.25	80.8	
.75	0	1:30	79.1	8	25	4M	3.06	37.4	52.5	Stable	7,000	0	+	+	+	+	
...	25	...	70.0	3.55	39.6	Stable	
2.0	0	1:15	80.5	8	20	4M, 3d	4.59	56.8	52.5	Stable	
...	18	...	73.4	1.12	12.9	
2.5	0	1:00	80.5	5	25	4M, 3d	3.16	39.2	53.5	3.232	1.268	.031	.20	130.0	Stable	37,000	0	+	+	+	+	
...	22	...	71.0	0.00	0.0	Stable	
4.25	0	12:30	79.0	8	22	3M, 4d	3.16	38.6	55.5	Stable	100,000	0	+	+	+	+	
...	22	...	71.8	0.0	0.0	Stable	
4.75	0	12:20	80.5	10	22	4M	4.47	55.4	59.5	2.152	.848	.040	.40	123.0	Stable	110,000	0	+	+	+	+	
...	20	...	73.1	8	38	3M, 4d	0.0	0.0	46.5	"	
5.25	0	11:30 A.M.	79.2	5	19	4M	4.31	52.6	77.5	1,450,000	+	+	+	+	+	
...	16	...	78.0	5	20	4M	2.01	24.3	55.5	870,000	0	+	+	+	+	
5.5	0	10:45 A.M.	78.5	5	18	4M, 3d	4.78	58.0	62.5	250,000	0	+	+	+	+	
5.75	0	10:40 A.M.	76.2	5	17	4M	3.96	47.0	57.5	Stable	2,100,000	+	+	+	+	+	
6.0	0	11:00 A.M.	78.1	8	16	4M	5.76	63.5	67.5	"	950,000	0	+	+	+	+	
6.5	0	3:45 P.M.	82.0	20	23	2M, 3v	5.95	74.9	68.5	2.280	.220	.060	.40	128.0	"	490,000	0	+	+	+	+	
8.0	0	4:10	82.0	8	29	3v	8.59	107.0	55.5	1.828	.172	.060	.35	119.0	"	300	0	0	+	+	+	
8.5	0	4:30	83.0	8	17	3v, 2d	8.93	113.0	77.5	1.668	.332	.060	.35	128.0	"	500	0	0	+	+	+	

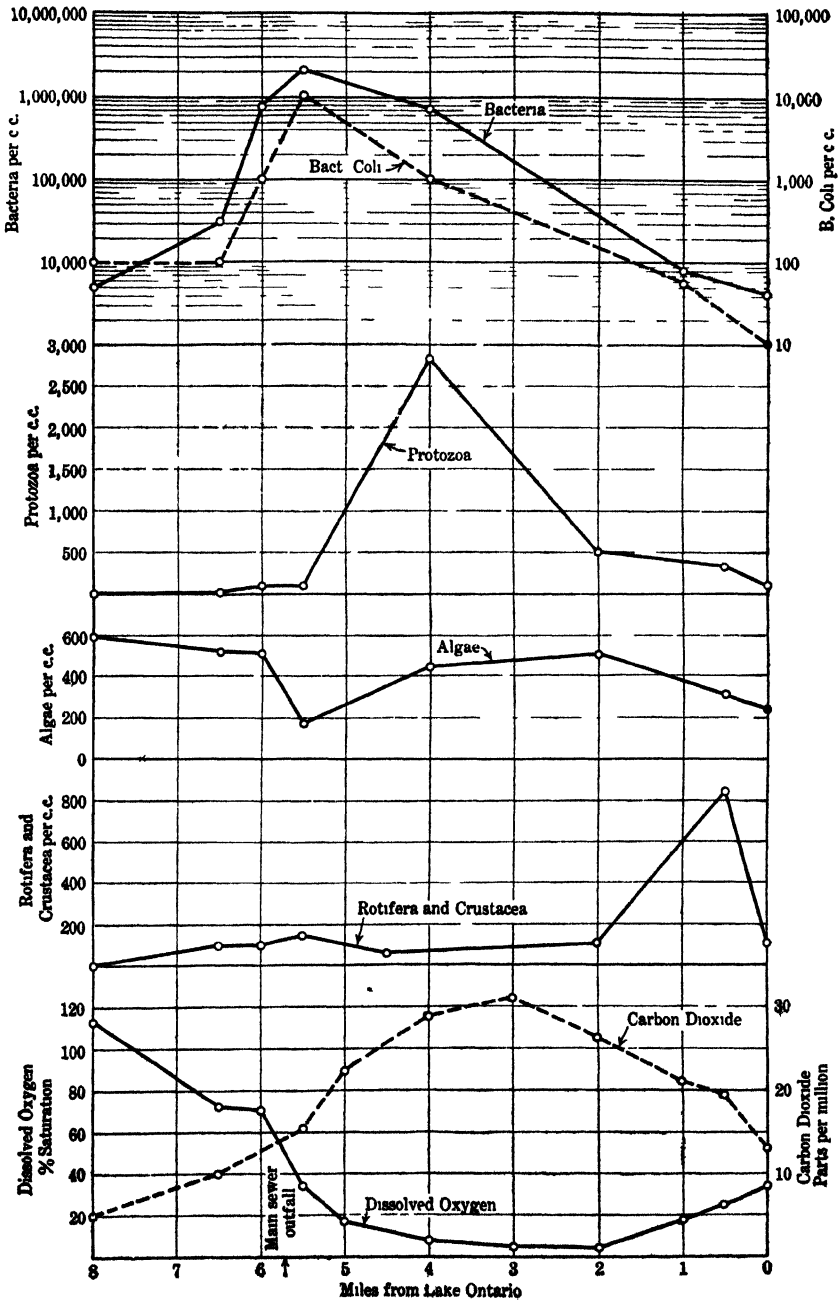


FIG. 92 — Changes in Dissolved Gases, Microscopic Organisms, and Bacteria.
Genesee River at Rochester, N. Y. July 24, 1912.

from the water. Another important factor is their destruction by larger microscopic organisms. These microscopic organisms, algæ, protozoa, rotifera, crustacea, etc., play an important part in the self-purification of the Genesee River during warm weather. A somewhat extended study of these organisms was made. Samples for microscopical examination were collected on different days at various points between the East Side Trunk Sewer and the lake at the surface, mid-depth and bottom. The variations in the relative numbers of the organisms of these different groups at various places in the river below the East Side Trunk Sewer illustrate in a typical way the biological action involved in the self-purification of a stream.

Immediately below the outlet of the trunk sewer there was a zone of heavy pollution within which the numbers of bacteria were very high, but the crustacea relatively low. Below this point of maximum bacterial life, the numbers of bacteria decreased to the lake. In the vicinity of the intense bacterial pollution and for a short distance below it, the numbers of protozoa were high, as might be expected from the fact that these organisms consume bacteria as food. At the same time there was a slight increase in the algæ, and their numbers were well maintained downstream to the river mouth. These vegetable cells utilize as food the oxidized products of the organic matter from the sewage. In the lower course of the river, for two or three miles back from the lake, the rotifera and crustacea made their appearance. They live upon the algæ and bacteria and especially upon the protozoa. Large numbers of crustacea were found in the lake hovering around the mouth of the river, waiting for the food that was being carried to them. Studies of these so-called plankton forms were made in the lake by the use of the plankton net, the results of which are given below. These crustacea serve as food for fish and that is why fish are attracted to the mouth of the river and why fishermen congregate along the breakwaters at the river mouth that extend about a third of a mile into the lake.

Speaking of the lake studies the report states:

Determinations of the microscopic organisms in the lake water were made on July 27th, August 2nd, and August 5th. In all of these samples diatoms and algæ were present in considerable variety. Generally speaking, these were more numerous at the surface than at the bottom, although certain species were occasionally found in great abundance at some intermediate point. In the samples collected near the shore, the number of microscopic organisms in the bottom samples varied according to the direction of the wind. When the wind caused the surface water to flow out at the bottom, the microscopic organisms were abundant in the bottom samples, but when the wind was off shore and cold, deep water being brought in at the bottom, the numbers of microscopic organisms in the bottom samples were lower. The microscopical examination of the surface-water along the beaches corroborated the temperature findings in indicating that with an off-shore wind the water at the shore line came from the bottom of the lake.

The protozoa that live upon bacteria and other microscopic organisms were most abundant in the immediate vicinity of the river mouth, and this was even true of the crustacea and rotifera. On August 15th a special study was made of the distribution of these larger organisms constituting the plankton, by the use of a plankton net. This net, kindly furnished by Prof. Charles Wright Dodge, was lowered to a depth of about 20 ft. and drawn to the surface at the rate of 1 ft. per second, in such a way as to collect the organisms present in about 100 gallons of water. The numbers of organisms found in the collected material were counted with the following approximate results:

TABLE 84
PLANKTON IN LAKE ONTARIO NEAR THE MOUTH OF THE POLLUTED GENESEE
RIVER

Samples from	Approximate Number of Organisms per Liter	
	Rotifera	Crustacea
Genesee River, opposite Naval Reserve Station.....	12	20
Genesee River, at mouth.....	285	23
Lake Ontario, $\frac{1}{2}$ mile from mouth of river ..	200	968
Lake Ontario, $\frac{1}{2}$ mile from mouth of river.	140	1035
Lake Ontario, 1 mile from mouth of river.	25	30

The variable character of pollution and self-purification near the river mouth is illustrated in Fig. 93. Here the distribution of bacteria in Lake Ontario indicates the zones of pollution established by different wind conditions. (See Chapter VII.)

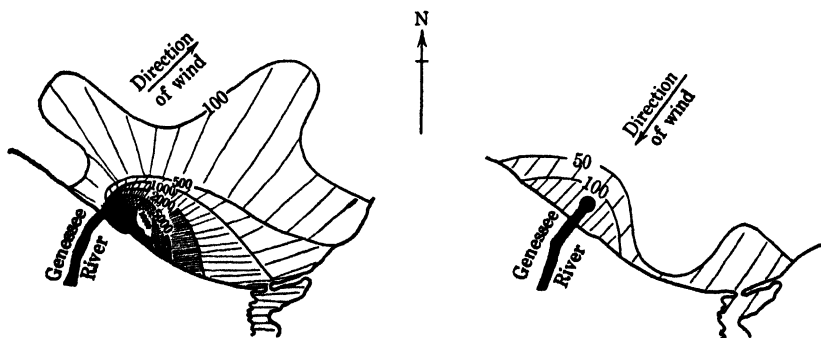


FIG. 93. — Pollution of Lake Ontario by the Genesee River as Indicated by the Numbers of Bacteria per cc. in the Lake Water.

Illinois River. — During the years 1909 to 1910 Forbes and Richardson made an extensive investigation of the Illinois River basin which receives the sewage from the City of Chicago. Purdy has summarized their report as follows:

Section 1. — Sanitary Canal at Lockport and Des Plaines River at Lockport. (This is about 33 miles below the source of the Sanitary Canal, which, after a flow of 38 miles, empties into the Des Plaines River at Joliet, Ill.)

Character. — Septic.

General and chemical conditions. — In canal, water clear. Offal, human excrement, and general refuse stranded or floating, but little decayed. Noticeable sewage odor. Sludge with foul odor. Dissolved oxygen averaged about 31.9 per cent of saturation. In Des Plaines River, water grayish. Decomposition well advanced. Foul privy odor. Conditions less offensive during cooler weather.

Biological content. — In canal, an occasional living fish. Many dead ones, floating or stranded along shore. (Fish were a common Lake Michigan species.) No animal life in bottom sludges. Scattered algae along edges of canal harbored the septic rotifer *R. actinurus* and the protozoa *Bodo*, *Vorticella*, *Oikomonas*, and *Anthophysa*, also septic. In Des Plaines River, bottom stones covered with sewage fungus (*Sphærotilus*) and blue-green algae. *Anthophysa*, *Carchesium*, *Paramecium*, and other septic protozoa abundant. Various less septic forms present. No fish in September, a few in November, but in dying condition. Many oligochaete and nematode worms in sludge. No mollusks or crustacea.

Section 2. — Des Plaines River at Dresden Heights (16 miles below mouth of the Sanitary Canal) and Illinois River from this point to Marseilles, a distance of 26 miles.

Character. — Polluted.

General and chemical conditions. — In Des Plaines River, water grayish, with privy odor. Dissolved oxygen averaged 13.1 per cent of saturation. In Illinois River, water grayish and clouded with tufts of sewage fungus (*Sphærotilus*). Masses of putrescent material floating on warmest days. Gas bubbles from bottom. Foul odor from both water and sludge. Dissolved oxygen ranged from 3.1 to 16.4 per cent of saturation. Unpolluted Kankakee (tributary) showed dissolved oxygen content of 108 to 127 per cent of saturation.

Biological content. — In Des Plaines River, large amounts of *Sphærotilus* and *Carchesium*. The blue-green algae *Oscillatoria* and *Phormidium* were frequent. Other algae present during cooler weather. No fish; no vertebrates except one frog. Many dead mollusks. Air-breathing beetles common. No sponges, hydroids, leeches, planarians, or crustaceans. In Illinois River, *Sphærotilus*, *Carchesium*, and *Vorticella* abundant. *Colpidium* and *Rotifer actinurus* present. Fish absent except an occasional specimen. Fungi and *Oscillatoria* in sludge; also immense numbers of worms (*Tubifex*). No mollusks, crayfish, or larvæ of may fly or dragon fly. Conditions a little better in cooler weather.

Section 3. — Illinois River from Marseilles (42 miles below mouth of Sanitary Canal) to Starved Rock, a stretch of 16 miles.

Character. — Contaminated.

General and chemical conditions. — Water grayish. Disagreeable odor. No gas bubbles. Dissolved oxygen about 16 per cent of saturation. Advent of Fox River (relatively pure) and passage of main stream over dam help to improve conditions at Starved Rock, where dissolved oxygen averages 43.6 per cent of saturation near north shore (due to Fox River) and 35.9 per cent near south shore.

Biological content. — *Sphærotilus*, *Carchesium*, *Epistylis*, and other septic organisms very few. At Starved Rock the only sewage organism found is small amounts of *Sphærotilus*. Fish in small numbers along edge of river, farther downstream are found in some variety in mid-channel, and at Starved Rock are moderately abundant. Blue-green algae (pollutional) fairly common near Marseilles, but scarce at Starved Rock, green algae increasing meantime. Some worms (*Tubifex*) in bottom mud. Insect larvæ, snails, crawfish, leeches, etc., more numerous.

Section 4. — Illinois River from Starved Rock (58 miles below mouth of Sanitary Canal) to Chillicothe, 107 miles below the canal. This section is 49 miles long.

Character. — Relatively pure water.

General and chemical conditions. — Water very slightly grayish, nearly normal; becomes greenish toward Chillicothe, and the slight odor of sewage disappears. Only in winter do bottom sludges show offensive odor. Dissolved oxygen 22 to 28 per cent of saturation in midsummer reaching maximum of 82 per cent in November.

Biological content. — Septic and pollutional organisms only occasional or absent. Fish numerous enough to justify commercial fishing. Green algae predominate, the blue-greens being only occasional. Water is made green by large amount of minute chlorophyll-bearing plankton. Large variety of insect larvæ and of mussels, snails, and crustacea. Sponges, leeches, bryozoa, and planarians are fairly common. *Tubifex* is rare in bottom sediments.

Ohio River. — The most extensive study of pollution and self-purification probably ever undertaken is that of the Ohio River. The work was done by the United States Public Health Service and has yielded a great amount of information on the mechanism of natural purification. Dealing with a very large and complicated river system with many and varied sources of pollution and widely separated sampling stations it is impossible to summarize the investigation for the purposes of this chapter. For this reason only typical examples of natural purification are chosen for illustrative purposes from the reports covering this study. Much of the material in these reports has already been drawn upon in preceding paragraphs of this chapter.

Bacteria. — The rate of bacterial self-purification of the Ohio River below Cincinnati is shown in Fig. 94. The way in which the lines approach straight lines on semi-logarithmic or ratio paper shows the analogy of self-purification to disinfection. As the environment is unfitted for development the organisms die off gradually. The more resistant forms or those best adapted to life under the conditions existing in the different reaches of the river survive longest.

Plankton. — The change in plankton content of the Ohio River below Pittsburgh is illustrated in Table 85. In general the sequence of growth and decline exhibited in this stretch of the river is similar to the findings in the Genesee.

Bottom Organisms. — The pollution of the Ohio River as indicated by organisms in bottom sediments is shown in Table 86. The effect of the discharge into the river of great amounts of sewage and industrial wastes from Pittsburgh, Cincinnati and Louisville is well illustrated.

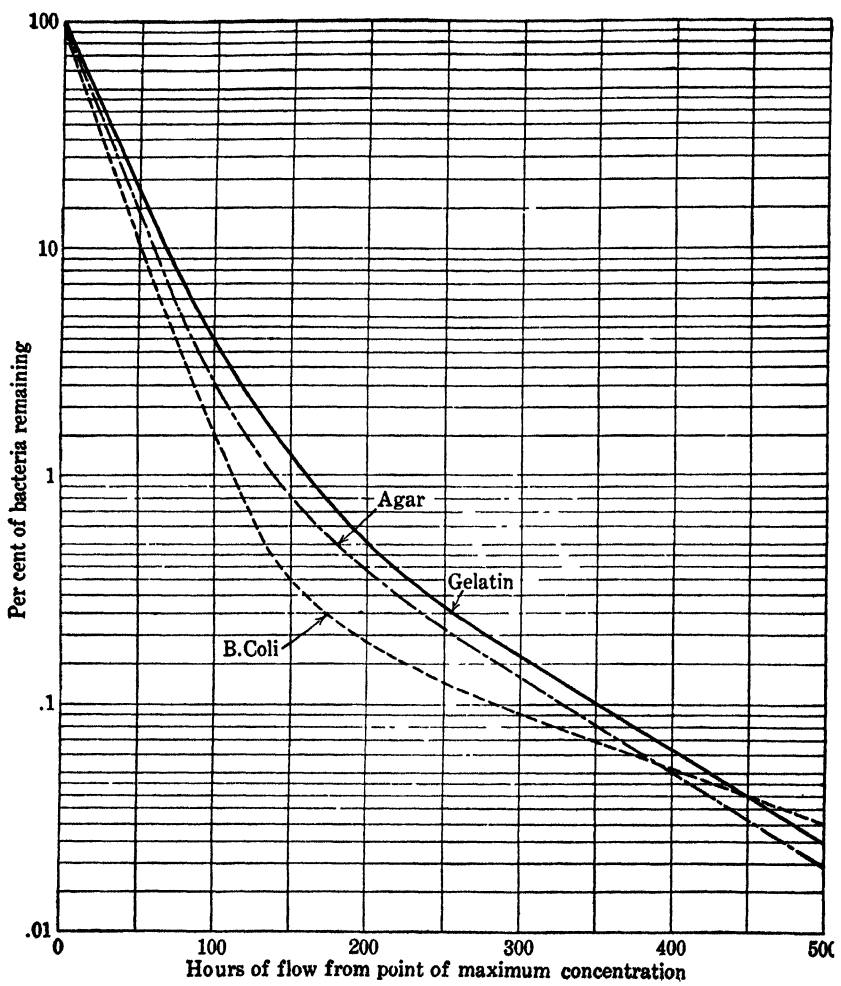


FIG. 94. — Rate of Decrease of Bacteria in the Ohio River between Cincinnati and Louisville. April to November, 1914–1916. After Frost and Streeter; U. S. Public Health Bulletin 143.

TABLE 85
PLANKTON IN THE OHIO RIVER SYSTEM ABOVE AND BELOW THE CONFLUENCE
OF THE ALLEGHENY AND MONONGAHELA AT PITTSBURGH

After Purdy

Average of 10 Samples, August to October, 1914
Organisms in Cubic Standard Units per cc.

Organisms	Monongahela River 12 Miles Above Confluence	Allegheny River 7 Miles Above Confluence	Ohio River	
			3 Miles Below Confluence	23 Miles Below Confluence
<i>Plants</i>				
Diatoms.....	3	40	8	54
Algae.....		20	15	18
Mastigophora.		13	1	7
<i>Animals</i>				
Ciliates.....	5	2	46	10
Rhizopods....	37	33	6	0
Rotifera	620	18	500	250
Crustacea.....		2	16	120
Vermes.....	8	6	14	1

NOTE: The Allegheny adds 60% to the volume of the Ohio, the Monongahela 40%.

TABLE 86
POLLUTION OF OHIO RIVER AS INDICATED BY ORGANISMS IN BOTTOM SEDIMENTS
After Purdy
Average number of organisms in 200 cc. of sample

Sampling Station				District	Pollutional Organisms	Doubtful or Indifferent Organisms
Monongahela	12 miles above	Pittsburgh		Pittsburgh	3	0
Allegheny	7 " " "				38	3
Ohio	3 " below "				582	1
349	" " "			Portsmouth	5	3
358	" " "				3	7
461	" " "			Cincinnati	1	2
475	" " "				91	2
492	" " "				12	5
598	" " "			Louisville	12	8
619	" " "				20	25
904	" " "			Paducah	3	3
920	" " "				0	3
933	" " "				0	1

Sangamon River. — In 1918 and 1919 a survey of the Sangamon River between Decatur and Springfield, Illinois, a distance of 120 miles, was made by Minna E. Jewell who has summarized the results of her biological and chemical findings as follows:

1. Plankton animals present varied greatly with season and water level. During the low waters of early fall only sewage and highly tolerant forms were found within 75 miles (by river) of Decatur.

2. The normal bottom fauna disappeared entirely below Decatur. Only typical sludge worms (tubificids) were found in the first 40 miles. The highly tolerant midge larva appeared within the next ten miles. Seventy-five miles below the source of pollution several of the normal bottom animals reappeared and about 10 miles farther down the normal bottom fauna was re-established.

3. Dissolved oxygen drops off, frequently to zero, below Decatur and at low water sufficient oxygen to support ordinary aquatic animals is not found again within 75 miles.

4. Oxygen consuming power of the water, organic nitrogen and ammonia rise rapidly immediately below Decatur and then gradually decrease to normal.

5. Dissolved oxygen, oxygen consumption, organic nitrogen and ammonia of the water fluctuate greatly with the temperature and volume of the stream.

6. Ammonia content of mud from the bottom of the river increases suddenly below Decatur and then decreases gradually to normal (about 80 miles below).

7. The ammonia content of the mud from any given location shows a seasonal variation due to temperature and oxygen content of the water. Slight temporary fluctuations in water level do not appreciably affect the ammonia content of the mud.

8. The worst conditions occur in the upper part of the river during hot dry summers, and in the lower part of the river in the early spring following a severe winter.

9. Chemical analyses of the water and mud afford the best criterion for the degree of pollution of the water at the time the sample is taken. The animals of the bottom form the best criterion for judging the worst conditions to which the location is subject.

The results of a biological survey of the Sangamon are tabulated in Table 87 (pp. 358-361).

Coweaset River. — The examples of self-purification so far considered have related to streams heavily polluted by crude sewage and other wastes. In 1914 and 1915 Weston and Turner investigated the digestion of a sewage-filter effluent at Brockton, Mass., by a small and otherwise unpolluted stream, the Coweaset River. They have listed the summary and conclusions of their study as follows:

1. The whole study reveals the extreme importance and the energetic action of certain plants and animals in purifying a stream receiving sewage effluents.

2. The Coweaset Stream, which receives a more or less nitrified sand-bed effluent, first lays down a pollution carpet by precipitation of organic and mineral matter, including silica.

3. This false bottom contains forms of life typical of pollution and purification which furnish a better index of the true condition of the stream than do organisms found in the supernatant liquid.

4. The entrance of organic matter into the stream increases plant and animal life in a series, beginning with the lowest and ending with the highest forms, each class appearing along with a definite food material.

5. Comparison of conditions on the Coweeset with investigations of other streams, shows that the smaller, shallower and more nearly stagnant the body of water, and the more highly nitrified and clarified the effluent, the more rapid is the succession of zones of higher animal life, and the more rapid and complete the purification process.

6. In this stream, when discharging less than 5,000,000 gallons and receiving 2,000,000 gallons of sand-bed effluent daily, the most important changes occur within the first mile; below which point, biologically speaking, the stream appears quite normal.

7. The return of the stream to a normal biological condition is more rapid than its return to a normal chemical condition; and the rate of chemical change is much less rapid below the point where biological processes have ceased their unusual activities.

8. It appears probable from the results of this investigation that a digestive process similar to that in the Coweeset River could be secured without difficulty for an effluent like that from the Brockton sewage disposal works by dispersing it throughout a shallow pond not more than four acres in area, before discharging it into the stream.

9. Natural storage reservoirs in a stream below the point of discharge of sewage effluent are of great assistance not merely in minimizing storm effects and seasonal changes but also in restoring the water to its normal condition.

10. In winter, indications of pollution extend farther down the stream than in summer. In other words, the critical or digesting region is elongated.

11. Although very slight changes are to be noted in winter, in summer, when biological activity is at its height, the 0.7 mile stretch of stream below Station 2, providing a normal storage period equivalent to about 1 hour's flow, effected, on an average, the removal daily of 39.5 lbs. of total nitrogen, 10.1 lbs. of nitrogen as albuminoid ammonia, and 101.3 lbs. of the carbonaceous organic matter as measured by the oxygen consumed test.

12. In this stream the dissolved oxygen tends to decrease until the excessive growth of animal life, favored by pollution, ceases. This factor outweighs both the normal reaeration of the stream and the oxygenating effect of plant growth.

13. The carbon dioxide introduced with the effluent tends to decrease downstream, at first rapidly and afterwards very slowly.

14. The rate of the oxidation of nitrogenous compounds varies with the season, and with the intensity of organic growth; it is very rapid in summer and hardly discoverable in winter.

15. The types of organisms which most clearly indicate the degree of pollution are bacteria, protozoa and diatoms.

16. Certain other types of higher organisms are characteristic of pollution, but in general the higher the type and the more varied its food supply the less reliable the form as an index of pollution.

17. The following organisms are commonly found in the polluted part of the stream:

Bacteria: Bact. coli and allied forms, Cladothrix.

Water Mold: Leptomitius.

Diatoms: Navicula forms, Gomphonema, Meridion.

TABLE
SELF-PURIFICATION OF THE
After

Date	Oct. 4, 1918	Oct. 5, 1918	Oct. 6, 1918	Oct. 7, 1918
Location	1 Mile Below Decatur	6 Miles Below Decatur	Seroggins Bridge, South of Harristown	South of Niantic
Estimated distance below Decatur by river, miles	1	6	20	30
Depth, feet.....	6	2½	3	3
T. ° C.....	18	18	18	16.5
D. O. p.p.m.....	Trace	0	0	0
per cent saturation...	0	0	0
Alkalinity p.p.m.....	300	260	348	286
pH.....	7	7	7.7	7.7
Appearance.....	Sewage	Inky	Inky, but opaque	Dark, slaty
Odor.....	Sewage	Putrid	Septic	Septic
Bottom organisms...		Heavy festoons of whitish gray moulds	Moldy slimes on bottom Oscillatoria along shore	Molds (unidentified) Sphærotilus natans Tubificid worms (appearing)
Plankton.....				

87

SANGAMON RIVER — 1918 TO 1919

Jewell

Oct. 8, 1918	Oct. 8, 1918	Oct. 11, 1918	Oct. 12, 1918	Oct. 13, 1918
Niantic Bridge	40 Miles Below Decatur	Bridge South of Illiopolis	3 Miles Below Illiopolis	West of Mt. Auburn 1 Mile Below Mosquito Creek
25	39	50	53	60
2	1	1	3	3
15	14.5	15.7	17	16
Trace	Trace	0.3	0.2	0
.....	3.0	2.0	0
286	320	300	310	300
7.6	7.6	7.8	7.9	7.9
Dark, slaty Septic	Dark, slaty Septic	Milky Less pronounced	Gray, turbid Disagreeable	Gray, turbid Not pronounced
Molds	Sphærotilus natans	Tubificid worms	Tubificid worms	Tubificid worms
Sphærotilus natans	Tubificid worms	Chironomid larvæ	Chironomid larvæ	Chironomid larvæ
Tubificid worms	Oscillatoria (along shore)	Nematode worms	1 crayfish near shore	Cambarus immunitus (crayfish)
	Nematode worms		Nematode worms	1 dead buffalo fish
	<i>Rotifers</i>	<i>Rotifers</i>	<i>Rotifers</i>	1 minnow apparently dying
	Rotifer citrinus	Rotifer citrinus	Rotifer citrinus	
	Rotifer neptunus	Philodina	<i>Ciliates</i>	Several small bull-heads gulping at surface, probably washed in from the creek
	Philodina	<i>Ciliates</i>	Vorticella	
	<i>Ciliates</i>	Carchesium	Colpidium	
	Oxytricha	Vorticella	<i>Flagellates</i>	
	Eschaneustyla	Colpidium	Euglena	<i>Rotifers</i>
	Carchesium	Epistylis	Phacus	Rotifer neptunus
	Vorticella	Paramecium putrinum	Anthophysa	Rotifer citrinus
	Colpidium	<i>Flagellates</i>	<i>Desmids</i>	<i>Ciliates</i>
	Paramecium putrinum	Euglena	Netrium	Carchesium
	<i>Flagellates</i>	Anthophysa vegetans	Pleurotænium	Vorticella
	Euglena	Chlamydomonas	Closterium	<i>Flagellates</i>
	Phacus	<i>Rhizopods</i>	<i>Filamentous algae</i>	Anthophysa
	Anthophysa vegetans	Arcella	Microthamnion	Euglena
	<i>Desmids</i>	<i>Diatoms</i>	Stigeoclonium	Phacus
	Pleurotænium	Navicula	Oscillatoria	<i>Desmids</i>
	Netrium	<i>Desmids</i>	<i>Molds</i>	Netrium
	<i>Filamentous algae</i>	Netrium	Unidentified	Closterium
	Stigeoclonium	Closterium	Sphærotilus natans	Pleurotænium
	Oscillatoria	<i>Filamentous algae</i>		<i>Filamentous algae</i>
	<i>Molds</i>	Microthamnion		Oscillatoria
	Unidentified	Stigeoclonium		<i>Molds</i>
	Sphærotilus natans	Oscillatoria		Unidentified
		<i>Molds</i>		Mold sygotes
		Unidentified		Sphærotilus natans
		Sphærotilus natans		

TABLE 87 —

Date	Oct. 14, 1918	July 20, 1919	July 20, 1919	July 20, 1919
Location	Frye Bridge North of Bolivia	Smith's Mill Above Dam	Above Roby Bridge, ¼ Mile Below Dam	3½ Miles Below Dam
Estimated distance below Decatur by river, miles.....	70	75	75	78
Depth, feet.....	6	6	1	2½
T. ° C.....	16	25	25	25
D. O. p.p.m.....	0	4.1	7 8	9 7
per cent saturation	0	48 9	93	115
Alkalinity p.p.m.....	320	287
pH.....	7.9	8	8
Appearance.....	Greenish gray	Green, slightly grayish	Green	Green
Odor.....	Faint	Noticeable	None	None
Bottom Organisms ..	Tubificid worms Chironomid larvæ	Tubificid worms Chironomid larvæ	<i>Crayfishes</i> Cambarus propinquus	<i>Crayfishes</i> Cambarus propinquus
Plankton.....	<i>Rotifers</i> Rotifer citrinus Philodina <i>Ciliates</i> Vorticella Paramecium caudatum <i>Flagellates</i> Euglena Phacus Anthophysa <i>Desmids</i> Netrium Closterium Pleurotæmium <i>Filamentous algae</i> Oscillatoria Microthamnion <i>Molds</i> Sphaerotilus natans Unidentified	<i>Crayfishes</i> Some carp said yet to be above dam <i>Rotifers</i> Rotifer citrinus <i>Ciliates</i> Vorticella Stentor Spirostomum <i>Flagellates</i> Euglena Phacus Eudorina <i>Desmids</i> Netrium Closterium Scenedesmus <i>Filamentous algae</i> Oscillatoria Spirogyra <i>Molds</i> Sphaerotilus natans	<i>Unionides</i> Quadrula undulata Quadrula lachrymansa Lampsis alata Lampsis luteola Lampsis anadontoides Unio gibbosus Anadonta grandis Tritogonia tuberculata Strophitus odentulus [tata Alasmodonta cosphærium <i>Gastropods</i> Pleurotera elevatum (a few) Campeloma sub-solidum <i>Insect nymphs</i> Caddis fly Heptagenia } May Hexagenia } flies Chironomid larvæ Tubificid worms in mud Fishing said to be fair for cat, carp and some game fish after the spring floods Plankton sample not taken	<i>Insect larvæ</i> Chironomid larvæ Heptagenia <i>Crustacea</i> 1 nauplius <i>Rotifers</i> Rotifer citrinus Notops <i>Ciliates</i> Several very small forms <i>Flagellates</i> Euglena Phacus Eudorina <i>Desmids</i> Netrium Closterium Scenedesmus Tetrademus <i>Diatoms</i> Stauroneis <i>Filamentous algae</i> Oscillatoria Spirogyra Tribonema

Continued

July 20, 1919	July 21, 1919	July 22, 1919
Northwest of Buckhart About 2½ Miles Below Robey	Riverton	Springfield Water Works
87	105	120
7	8	15
25	26.2	27
12.8	7.5	9.8
152	91	109
....
....	8
Green	Green	Green
None	None	None
<i>Unionida</i>	<i>Unionida</i>	Bottom conditions abnormal because of dam
<i>Lampsilis luteola</i>	<i>Lampsilis luteola</i>	<i>Rotifers</i>
<i>Lampsilis alta</i>	<i>Lampsilis alata</i>	<i>Notops</i>
<i>Quadrula pustulosa</i>	<i>Unio gibbosus</i>	<i>Philodina</i>
<i>Quadrula undulata</i>	<i>Quadrula undulata</i>	<i>Ciliates</i>
<i>Tritigonia tuberculata</i>	<i>Quadrula pustulosa</i>	A few small forms
<i>Symphynota costata</i>	<i>Tritigonia tuberculata</i>	<i>Paramecium</i>
<i>Fish</i>	<i>Anadonta grandis</i>	<i>Flagellates</i>
Carp } seen	<i>Sphaeriidae</i>	<i>Euglena</i>
Bullhead } seen	<i>Sphaerium</i>	<i>Phacus</i>
Channel cat, reported by fish- ermen	<i>Musculium</i>	<i>Eudorina</i>
<i>Insect larvae</i>	<i>Gastropods</i>	<i>Pandorina</i>
Caddis worm (a few)	<i>Pleurocera elevatum</i>	<i>Glenodinium</i>
1 <i>Corydalis</i> larva (Helgram mite)	<i>Campeloma subsolidum</i>	<i>Rhizopods</i>
	<i>Insect larvae</i>	<i>Arcella</i>
	May fly nymphs (<i>Hexagenia</i>)	<i>Desmids</i>
	Chironomid larvae	<i>Netrium</i>
	<i>Fish</i>	<i>Closterium</i>
	Sunfish (small) observed	<i>Scenedesmus</i>
	Bullhead } Reported as common	<i>Tetrademus</i>
	Carp } in spring	<i>Diatoms</i>
	Buffalo }	<i>Fragillaria</i>
	Cat }	<i>Filamentous algae</i>
		<i>Oscillatoria</i>
		<i>Tribonema</i>
		Unidentified fragments

Protococcales: Protococcus.

Desmids: Closterium.

Filamentous Algae: Spirogyra communis, Tribonema (Conferva).

Phanerogams: Potamogeton, Sparganium, Sagittaria, Polygonum, Glyceria.

Protozoa: Amœba, Arcella, Chlamydomonas, Euglena viridis, Monas, Carchesium, Colpidium, Euplotes, Lionotus, Loxophyllum, Metopus, Oxytricha, Vorticella.

Bristle Worms: Nais, Tubifex.

Rotifers: Rotifer, Diglena.

Leeches: Erpobdella punctata, Glossiphonia stagnalis.

Snails: Planorbis trivolvis, Physa heterostrophæ.

Crustacea: Asellus communis, Cyclops, Pleuroxus, Simocephalus.

Insect Larvæ: Chironomus decorus, Ptychoptera.

18. Most of the organisms occur in "blooms" which are subject to fairly regular seasonal variations.

19. Chemical changes in the stream are due to bacteria more than to any other group of organisms. The higher animals are important by removing an excess of lower organisms.

20. The rate of decrease of bacteria is most rapid at the point of entrance of the effluents, and except during the winter floods very slow below the first mile.

21. Diatoms are more abundant in the normal stream, but certain forms can live in highly polluted water.

22. The algae are increased in quantity by the entrance of the effluent. Certain forms are shown to have a preference for polluted and others for unpolluted water.

23. The water mold, Leptomitus, has been found when oxygen was plentiful and the alkali- and nitrate-producing bacteria fewer and less active.

24. Different species of higher plants will tolerate pollution in different degrees. They are important in holding back suspended matter and preventing small organisms from being washed down by the current; they also assist in the removal of colloidal suspended matter and in checking the flow of the stream.

25. Abundance of Hydra is apparently correlated with the abundance of small crustacea.

26. Large numbers of rotifers and small crustacea may indicate the presence of simple green algae and not necessarily of sewage pollution.

27. Certain worms and insect larvæ assist materially in the digestion of the débris accumulated in the pollution carpet.

28. Fishes (trout, suckers) were occasionally observed, even in the most polluted part of the stream.

The yearly averages of physical, chemical, and biological data secured along the stream are shown in Fig. 95.

Biology of Sewage Treatment. — The forces of self-purification of water, physical, chemical and biological, are called into great activity in many processes of sewage treatment. Towards the end of the last century sewage treatment methods were introduced that relied largely upon biological activity to transform the unstable organic matter contained in sewage into more stable compounds that did not create a nuisance in the streams into which the treated sewage was finally discharged. The newer methods of sewage treatment have continued to employ biological forces of purification. Modern sewage treatment,

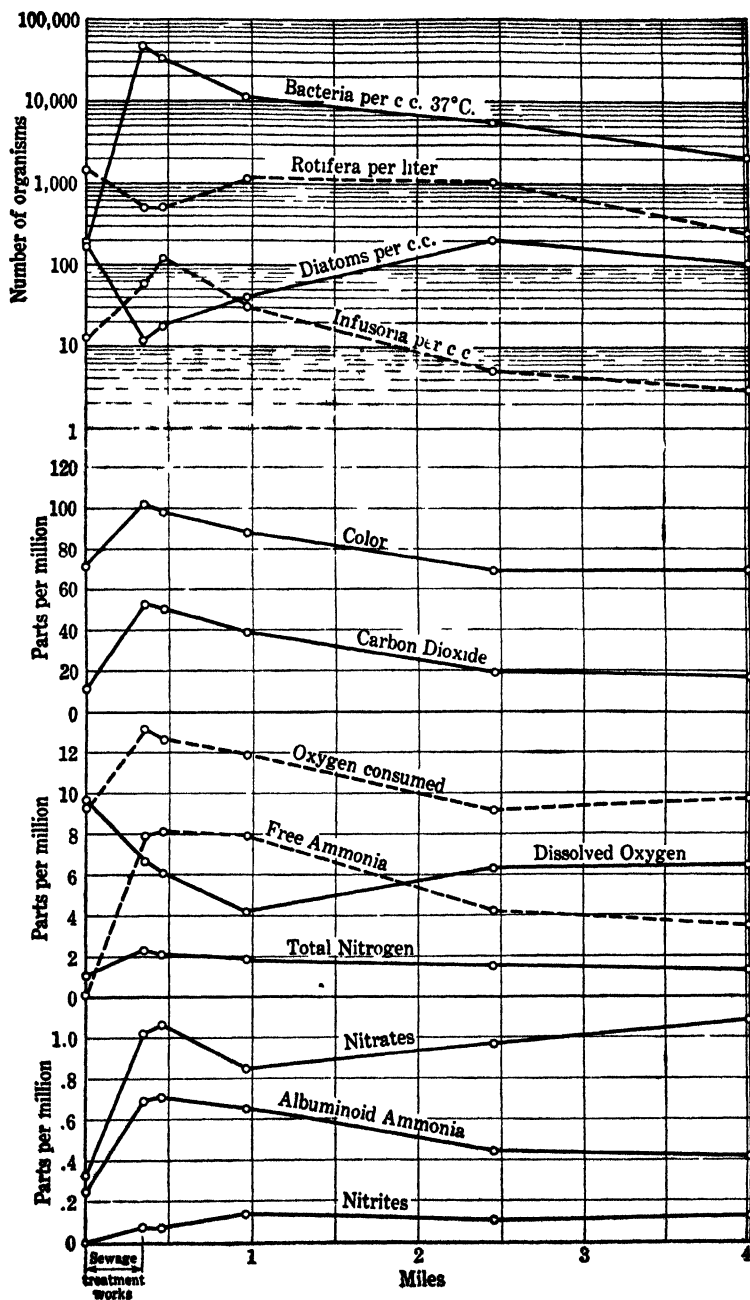


FIG. 95. — Self-Purification of the Coweaset River below Brockton Sewage Filters. After Weston and Turner.

indeed, is in many ways analogous to the self-purification of water. Purification activity, however, is intensified and many of the reactions that proceed slowly in bodies of water that receive sewage are accomplished more quickly in sewage treatment works. It is the function of biological sewage treatment methods to create environmental conditions that induce or stimulate the various forces of purification that operate under the misnomer of "laws of chance" in Nature's household.

A discussion of the biology of sewage treatment falls outside the province of this book. Suffice it to state that the principles of sanitary microscopy discussed in this and other chapters are applicable to sewage treatment and disposal, as well as to collection, storage and purification of water. The organisms encountered are the same or very similar. The life of Imhoff tanks for example is much like that found in the septic portions of the zone of active decomposition, and the organisms working in sprinkling filters or activated sludge tanks are equally industrious in the mesosaprobic stretches of polluted streams. Sewage treatment has received a tremendous impetus since engineers have learnt to imitate Nature or better to utilize those natural forces that have been evolved in Nature in order to create a balance of life, without which life itself would soon become extinct. In sanitary science as in some other fields of knowledge "Nature is the best teacher."

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CHAPTER XIII

CONTROL OF ALGÆ

The best way to eliminate odors and tastes produced by microscopic organisms, more particularly in lakes, ponds, reservoirs and other standing bodies of water is to control the growth of the plankton *in situ*. Numerous methods have been devised in the attempt to accomplish this control. Some of them try to do so by reducing the available food supply or by so modifying the chemical composition of the water that it will not support large growths of plankton; others provide for poisoning the organisms by the addition of chemicals to the water; still others are based on the control of some of the physical factors that affect microscopic growths. All of these methods have their uses and are applicable in one way or another under due consideration of the controlling local conditions.

PREVENTION OF ALGAL GROWTHS BY SUITABLE RESERVOIR CONSTRUCTION AND OPERATION

The basic sanitary requirements for the storage of water and its maintenance in a satisfactory condition have been set forth in Chapter X. The effects of various agencies on plankton growth were there discussed. In the following the methods available to reduce or prevent algal troubles are brought together.

Preparation of Catchment Area. — Since the quality of water reaching lakes and reservoirs is determined by the characteristics of the catchment area from which the water is collected, much can be accomplished by suitable preparation of the collecting grounds. In general the catchment area should be so conditioned that the water falling upon the clean portions will not pass through portions rich in organic matter before reaching the reservoir. Preparation of the catchment area usually takes the form of treatment of swamps, ponds and other areas of sluggish flow in the courses of brooks and streams, in which the water may pick up plankton food, or from which spores and vegetative plankton may be carried into the storage basins that serve as sources of water supplies.

Swamp treatment in particular yields good results. We may distinguish between three types of swamps: (1) Rainwater swamps which

are due to the accumulation of rain water or the overflowing of streams during floods; (2) Backwater swamps which are areas of shallow flowage formed in sluggish streams, particularly at bends or other obstructions to flow; (3) Seepage outcrop swamps which usually occur at the foot of hillsides where there is an outcrop of a stratum of water-bearing sand or gravel underlain by a stratum of clay, shale or other impervious material. Of these the seepage outcrop swamps are probably the most troublesome. They are less subject to desiccation during drought and since much ground water accumulates in them they are apt to contain relatively large amounts of carbon dioxide and nitrates which are aids to the growth of all aquatic vegetation.

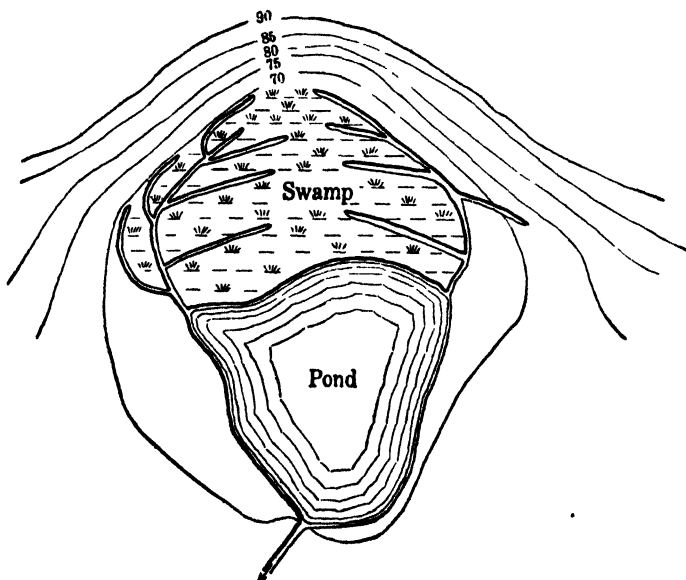


Fig. 96. — Drainage of Seepage Outcrop Swamp.

The method of swamp drainage to be used depends upon the type of swamp. In the first, the rain water swamp, ditches should be provided to carry off the surplus water after the main flood waters have passed. In the second, the stream channel should be regulated to lower the water in the swamp and increase the flow. In the third, a system of marginal drains should be constructed to intercept the seepage water; central drains must sometimes be added to assist in lowering the ground water table below the surface of the ground. The treatment of a seepage outcrop is illustrated in Fig. 96.

Reservoir Construction. — A further opportunity to control the growth of microscopic organisms which, it has been shown, is favored by certain conditions of under-water topography and geology is afforded at the time of reservoir construction by the proper preparation of the reservoir site before the water is impounded. In natural ponds and lakes, too, there are certain operations such as swamp treatment and shore line clearing that can be undertaken to good advantage.

Treatment of Ashokan Reservoir. — In 1907 Hazen and Fuller made the following recommendations for the conditioning of the Ashokan Reservoir of the New York Water Supply System.

1. *Clearing and Grubbing.* — Cut all trees and bushes close to the ground over the entire area of the sides and bottom.

2. *Burning Vegetation.* — Burn all grass, weeds and shrubs and see that this is done shortly before the areas are flooded. In other words, do not allow the water to flood any areas on which expansive growths of weeds have occurred since the original preparation of the area.

3. *Preparing the Shores.* — Around the shores to a vertical depth of at least 20 ft. below high water, remove all stumps, and, so far as necessary, roots and other matters which might become exposed by continued wave action, and leave the surface with even slopes, so that the shores will be maintained in a presentable condition when the water is drawn down. We do not think it necessary to spend a large amount of money in this preparation. The wave action will tend to clean it and accomplish the desired results, but some extra attention should be given to it with reference to its appearance when exposed, and also to prevent, as far as possible, the leaving of enclosed shallow areas which might serve as places where the spores of organisms would remain and serve as centers of infection, when conditions in the reservoir became favorable.

4. *Preparing the Bottom.* — After removing all the top vegetation from the swamp areas, which can be done by cutting it off close to the surface and burning, careful examination should be made for places where the surface "crust" is so loosely attached to underlying soft material that it might rise after the reservoir is full. We have given this question some attention when examining the swamps, and their surfaces wherever we have seen them are such that this factor does not appear to be of much importance here. However, experience elsewhere indicates that it should be given further attention.

The reasons for Hazen and Fuller's recommendations are either self-evident or explained in the above paragraphs. The recommendations aim first at the removal of that portion of the vegetation which is readily cleared and which presents the bulk of the organic matter in the areas to be flooded; next at the conditioning of the shores that become exposed as the reservoir is drawn down so that they may not become unsightly; last, at the treatment of swamps and areas of shallow flowage which may either affect the physical and chemical condition of the water while submerged or serve as breeding grounds for microorganisms when the reservoir is drawn down and shallow

pockets are sufficiently exposed to permit the development of swamp-like vegetation.

Treatment of Swamps, Pockets and Areas of Shallow Flowage in Reservoirs. — The treatment of swamps, pockets, and areas of shallow flowage has frequently been carried further than suggested for Ashokan Reservoir. The methods of dealing with swamps from which water courses feed into the reservoir have already been discussed. Existing marginal swamps can be treated in much the same way. Swamps that become submerged when streams are impounded to form storage reservoirs, however, require different conditioning. The usual method is to remove the swamp muck entirely from the reservoir site or, if the muck is found to be very deep or far submerged, to cover it with a layer of clean sand and gravel.

The construction of reservoirs sometimes results in the creation of swamps in the areas of shallow flowage so frequently found at the head of reservoirs. Such areas can be deepened by excavation or, as in the case of the Scituate Reservoir at Providence, the water level can be raised by building at the upper end of the reservoir a control dam which will serve the purpose of keeping the water from being lowered in the shallow reaches during the seasons of plant growth. Pockets that result in the formation of stagnant pools when the reservoir is drawn down should be made self-draining by the construction of proper ditches.

Old reservoirs that have given trouble due to the existence of swamps and shallow flowage in the reservoir area can be improved by methods similar to those just outlined. Thus reservoirs No. 2 and 3 of the Boston Water Works were reconditioned after construction by increasing the depths at points of shallow flowage by excavation and by removing all stumps and much of the muck from the sides and bottom as far as they could be exposed by lowering the water level. The upper reaches of Lake Cochituate were treated similarly as a result of complaints that areas of shallow flowage became offensive mud flats when the lake was drawn down during the summer months.

Soil Stripping. — The methods of algæ control so far discussed have dealt only with the removal of the grosser portions of the organic matter that may cause trouble in the storage of water. There remain two other sources of organic matter, the top soil or layer of living earth and the water itself. The elimination of organic matter by the removal of the top soil from the reservoir site is known as *soil stripping*. The clarification by sedimentation of the water entering the reservoir is called *pre-storage*.

Soil stripping has been a Massachusetts custom. There are but few examples of this practice in other parts of the world. In Europe only

a limited number of impounding reservoirs have been stripped. These are small and were built on peaty areas. Stripping was apparently undertaken for the reason that at lower points on the same stream older reservoirs were found to have given trouble for a time in earlier years. One of these stripped reservoirs is near Bradford, England.

In Massachusetts the cleaning of the bottoms and sides of reservoirs was begun about 1883 following the occurrence of seriously objectionable tastes in the Boston reservoirs during the unusually dry season of 1881 to 1882. At that time the more recently constructed storage reservoirs of Boston had had trees and brush growing on the bottom and sides cut down and removed or burned. Shallow flowage had also been eliminated to some extent. As trouble was still encountered Reservoir No. 4 of the Boston Water Works, known as the Ashland Reservoir, built in 1882 to 1885 was treated even further. The bottom and sides were thoroughly cleaned of all loam, stumps and vegetable matter and the reservoir was deepened wherever the original depth below high water was less than 8 feet. The later reservoirs of the Metropolitan Water Supply System, as well as those of some other cities and towns, such as Worcester and Cambridge, were similarly treated.

Results of Stripping in Massachusetts.— In 1907 Hazen and Fuller summarized as follows the results of observations made by the Massachusetts State Board of Health upon the odors and tastes encountered in stripped and unstripped reservoirs. (Table 88.)

Hazen and Fuller concluded as follows:

These results indicate a substantial reduction in odor in the stripped reservoirs and the reduction is no doubt largely due to stripping.

The chief fact, however, to be learned from the practical application of reservoir stripping in Massachusetts is that it does not entirely or uniformly eliminate unpleasant or offensive odors from impounded surface waters. This is shown by occasional tastes and odors even in the Ashland, Hopkinton and Wachusett reservoirs as they continue in service. It certainly reduces these odors to a considerable extent when compared with the results obtained under more or less comparable conditions from unstripped reservoirs. But the evidence is clear that stripping alone cannot be relied upon to produce an impounded water satisfactory as to tastes and odors at all times.

Progressive Improvement of Stripped and Unstripped Reservoirs.— When a reservoir first fills with water, the organic matter left on the reservoir bed gradually decomposes, and the water becomes colored with vegetable stain and absorbs many substances that favor plankton growth. As time elapses, however, the amount of organic matter available from this source decreases and with it the color and the microscopic life. A point is finally reached when a marked reduction no longer occurs from year to year and the reservoir seems to have reached

TABLE 88

COMPARISON OF ODORS IN STRIPPED AND UNSTRIPPED RESERVOIRS
Massachusetts Experience with Reservoirs over 100 Acres in Area

Stripped Reservoirs

Place	Reservoir	Year Put in Service	Area, Acres	Capacity, M.G.	Average Depth, Feet	Supply Held, Days	Odor Group
Worcester	Lower Holden	149	742	15.2	161	I
"	Kent.....	119	513	13.2	142	II
"	Upper Holden	185	794	16.8	174	II
"	Leicester.....	143	681	14.6	235	II
Met. Water	Sudbury.....	1897	1292	7,253	18	332	II
District	Wachusett....	1905	4200	63,100	46	534	III
"	Framingham 2	1878	134	530	12	12	III
"	Framingham 3	1878	253	1,183	15	43	III
"	Ashland.....	1885	167	1,464	26	227	III
"	Hopkinton....	1894	185	1,521	26	261	III
Cambridge	Lower Hobbs	467	1,450	10	220	III

Average odor group..... 2.5

Unstripped Reservoirs

Holyoke	Wright & Ashley.....	280	1510	16	500	III
Holyoke	Whiting.....	115	500	13	300	IV
New Bedford	Old Storage...	300	400	4	550	IV
Lynn	Walden.....	128	403	12	308	V
Springfield	Ludlow.....	387	1344	11	75	V
Whitman	Hobart's Pond	175	V

Average odor group..... 4.3

DESCRIPTION OF ODOR GROUPS

Group I. Waters which are odorless or which have occasional faint odors.

Group II. Waters which are usually odorless but have occasionally a distinct and at times an unpleasant odor.

Group III. Waters which have frequently a noticeable and at times a distinct or unpleasant odor.

Group IV. Waters which have generally a noticeable odor which is frequently unpleasant or disagreeable.

Group V. Waters which have generally a strong and frequently an unpleasant or disagreeable odor.

a state of normal color and normal plankton growth. It is prior to this time that the differences between the various types of impounded reservoirs — stripped, unstripped, swampy, and clean — must be expected to be especially marked. As shown in Fig. 97 a study of the changes in color and microscopic growths that occur during the years following the first filling of impounded reservoirs indeed yields much valuable information on the merits of soil stripping and swamp treatment. The information presented in this figure was compiled by Mr. X. H. Goodnough, Chief Engineer of the Massachusetts State Department of Health, and is here shown by his courtesy. The color changes are given in order to indicate the variation in the organic content of the water. For purposes of comparison the reservoirs studied are divided into five classes which array themselves in order of the amount of initial color observed in their waters as follows:

1. Large deep reservoirs — uncleaned, flooding swamps.
2. Shallow reservoirs — uncleaned, flooding swamps.
3. Large deep reservoirs — uncleaned, but not flooding extensive swamps.
4. Large deep reservoirs — cleaned.
5. Small reservoirs — cleaned.

During the first decade after filling, color reduction takes place rapidly; after that more slowly until a condition of stability seems to be reached. In general the reservoirs maintain throughout the years of record the same relative position with respect to color that they occupied when they were put in service. After about twelve years, however, the color in all reservoirs, but those in classes 1 and 2, has been lowered to substantially the same value. The extraction of organic matter from the soil seems to have been completed while the swamp muck is still yielding coloring matter. In this connection it is interesting to know that clayey soil gives up its organic content far more slowly than sand or gravel, while peat, which is about 60 per cent organic, yields very little coloring material.

The progressive decrease in microscopic organisms is not as regular as the reduction in color. This is not unexpected, considering the many factors other than the presence of food materials that affect plankton growths. The microscopic counts obtained from year to year, however, manifest a distinct trend from relatively high to moderate values. There is a general correlation between the decrease in color and the reduction in microscopic growths. The classes remain in the same order and the presence or absence of swamps is particularly noticeable. As in the case of odors and tastes the effect of soil stripping

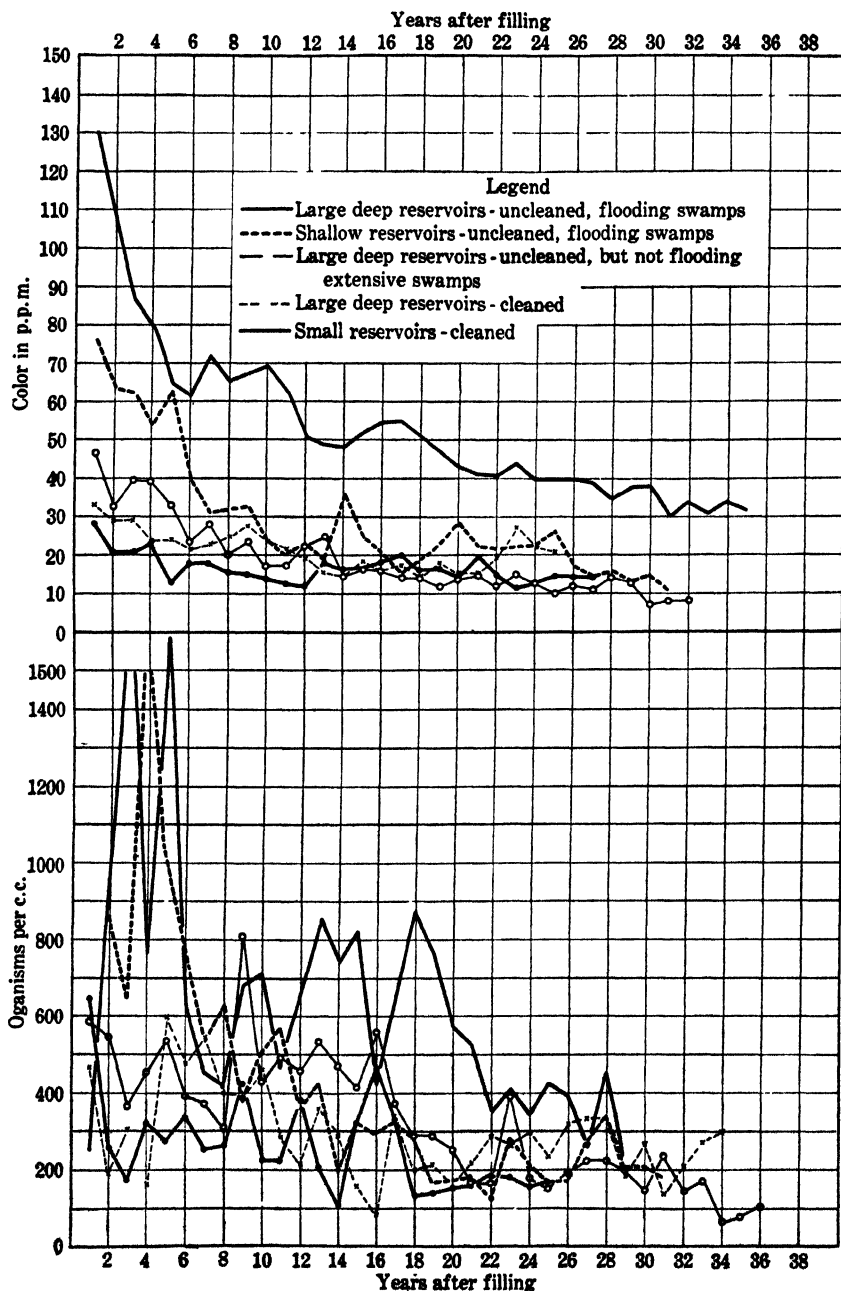


FIG. 97. — Progressive Reduction in Color and Microscopic Organisms of Massachusetts Impounding Reservoirs Subsequent to their Filling. *Courtesy of X. H. Goodnough, Chief Engineer, Massachusetts State Department of Health.*

upon color and organisms is marked, especially so during the first years of service. This suggests the importance of comparative studies of reservoir cleansing *versus* water purification wherever impounded waters are to be used. Whatever method is cheaper relative to the results obtainable should be employed.

Pre-Storage. — As pointed out in Chapter X, reservoirs that were clean when they were put into service do not stay so indefinitely. If it were possible to collect from the catchment area water that contained no foreign substances, deposits would not form on the reservoir bed. Since this is not the case sedimentation and the growth and death of microscopic organisms will result in the gradual accumulation of bottom deposits that sooner or later assume a controlling part in the reservoir characteristics. It is possible, however, to limit the amounts of foreign materials carried into reservoirs by providing small basins near

TABLE 89
RESULTS OF PRE-STORAGE
After Houston

Description of Sample	No. of Samples		No. of Bacteria per cc. (Agar 37° C.)	
	Chemical	Bacteriological		
Inlet to Hornsey Reservoir.....	47	52	92.2	
Outlet from Hornsey Reservoir.....	47	52	56.0	
Per cent Improvement..			39.2	

Description of Sample	Parts per Million				Color*
	Free Ammonia	Albuminoid Ammonia	Oxygen Consumed	Turbidity	
Inlet to Hornsey Reservoir.....	.030	.093	.750	13.3	34
Outlet from Hornsey Reservoir.....	.024	.089	.708	10.4	33
Per cent Improvement..	20.0	4.3	5.6	21.8	2.9

* Mm. brown in 2 ft. tube.

the head of main reservoirs or by damming off the arms or branches through which the incoming water passes and in which it deposits part of its load before reaching the main basin. Houston has called this "pre-storage" and has shown that marked improvement can be expected by providing basins with a capacity of 15 to 16 hours' storage. (See Table 89.) The use of pre-storage basins depends naturally upon local facilities for their construction, by-passing and cleaning. It is probably more easily feasible where large artificial reservoirs are to be used than where impounded supplies are to be created.

Reservoir Operation. — The reservoir once constructed, plankton troubles can often be reduced by intelligent operation and proper use of the intake facilities that are commonly provided at gate houses. This is also true of water supplies derived from natural lakes and ponds.

By-passing Troublesome Reservoirs. — A method of control readily used is to shut off or by-pass the troublesome reservoir when it is possible to do so. In three weeks to three months the growths usually disappear. Sometimes, however, growths will last longer than is commonly required for the reservoir to "work itself off." Thus Hale reports that *Asterionella* appeared in Ashokan Reservoir, New York City, in quantity in November, 1919, and lasted until March, 1920. *Tabellaria* prevailed in Kensico Reservoir from February to June, 1919, and *Aphanizomenon* from June to September, 1919, reaching a maximum of 5000 units. *Dinobryon* was present in Silver Lake reservoir, Staten Island, for many months.

When it is necessary to put the reservoir back into service quickly, the water can be treated with copper sulphate in order to hasten the removal of the plankton growths. If the organisms are killed while the reservoir remains in operation, tastes and odors due to decay and to liberation of essential oils are apt to be annoying. This is particularly so with distributing reservoirs from which the water flows directly to the consumer. By-passes are, therefore, an essential equipment of uncovered distributing reservoirs. When by-pass and intake facilities so permit, it is sometimes possible to close off only that part of the reservoir which is giving trouble. This is the case with Ashokan Reservoir, which is constructed to form two independently controlled basins.

Shifting Depth of Draft. — A method of control frequently overlooked by water works operators and of almost universal application is found in shifting the draft from one depth to another. According to Hale the policy of deep draft was adopted at Croton Lake in 1912 following two years of complaints caused by the growth of *Aphanizomenon*. By shifting the draft to a depth of 75 feet a reduction in microscopic organisms of 65 to 75 per cent was obtained as shown in the following table:

TABLE 90
REDUCTION IN ORGANISMS BY SHIFTING DRAFT
Croton Supply, third quarter — July, August, September
After Hale

Year	Temperature	Microscopic Organisms	Reduction
1911	72° F.	2836	
1912	67°	1072	65%
1913	65°	677	75%

A knowledge of the mode of occurrence of the various plankton species makes it possible to predict the depth of draft that will yield the best results during different seasons. The reader is referred to Chapter IX in which seasonal occurrence and vertical distribution of organisms have been discussed. Laboratory control, however, is essential when greatest efficiency is desired.

Benefits other than the reduction in plankton can also be secured by shifting the draft. Thus in the above example the temperature of the water as sent to New York City from Croton Reservoir during the summer was lowered by 5° to 10° F. Besides plankton and temperature, color, turbidity, iron, manganese, carbon dioxide, oxygen and gases of decomposition all vary with depth, and their presence in the water drawn can be controlled to suit the conditions desired, provided gate houses are equipped with proper intake facilities. The more flexible a reservoir system is made by provision of by-passes, multiple intake gates, and other operating equipment, the fewer will be the unavoidable troubles. Engineers should bear this in mind in the design of storage and distribution systems.

Control of Water Weeds. — In properly constructed reservoirs the growth of water weeds is usually limited, although great masses of higher aquatic plants are known to have occurred in clean but shallow lakes, ponds and reservoirs. There are four ways of removing these aquatic growths — *draining, dredging, cutting and dragging*. If a reservoir can be *drained* during hot weather the exposed aquatic plants will rapidly die and their roots will be killed. This, however, means throwing the reservoir out of service at a time when it is usually most needed. If the reservoir is drawn down in the fall, the roots will not dry and must be destroyed separately. There is also danger of propagating weeds by seeding the exposed bottoms. *Dredging* can be undertaken at any season and will remove the roots of the plants as well as the stems and

leaves. *Cutting* offers at best a temporary means of relief. In shallow water, plants can be cut by a scythe; in deeper water a flexible saw similar to that shown in Fig. 98 must be used. This saw is operated from two boats. It is weighted with sinkers so that it will lie close to the bottom and is pulled back and forth to cut the weeds. The cut weeds

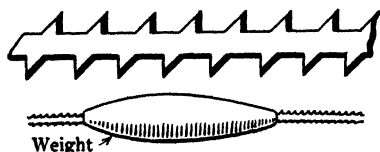


FIG. 98. — Flexible Saw for Cutting Subaqueous Weeds. After *Engineering News-Record*, Vol. 95, 1925, p. 638.

float to the surface and must be removed, as they will otherwise decay and adversely affect the quality of the water. In shallow places and along the shore both the foliage and roots of weeds can be removed by *dragging* along the bottom steel cables equipped with steel clips and swivels.

Arsenical compounds such as sodium arsenite will destroy a variety of water weeds. Their use in lakes, ponds and reservoirs that serve as municipal water supplies is, however, out of the question.

Reservoirs for Filtered Water and Ground Water. — The principles of reservoir construction and operation outlined in the preceding paragraphs apply particularly to impounding reservoirs, natural lakes and ponds and large distributing reservoirs for the storage of unpurified surface water. When filtered water or ground water is to be stored, certain additional safeguards against the growth of algæ should be provided.

The most important safeguard is the exclusion of light. Darkness deprives the chlorophyll-bearing plankton organisms of energy for growth, and, since there are but few other troublesome organisms, complaints arising from the presence of algæ are commonly eliminated by storing clean water in the dark. Contrary to public opinion, the water does not deteriorate during storage. The differences in microscopic growths in covered and uncovered reservoirs are illustrated in Table 91 which shows the prevalence of microscopic organisms in samples of water from the low and high service reservoirs in Ancon, C. Z. These reservoirs are supplied with filtered water from the Miraflores water plant.

Besides eliminating microscopic organisms, the construction of covered reservoirs has additional advantages. The water is kept at a more uniform temperature, colder in summer and warmer in winter, making it more palatable and reducing the danger of freezing both hydrants and service and plumbing pipes. Atmospheric dust and dirt are kept out of the water, and birds, animals and human beings have no access to it. The sanitary advantages of storing filtered water in covered

TABLE 91

MICROSCOPIC ORGANISMS IN COVERED AND UNCOVERED RESERVOIRS, ANCON, C. Z.
After Bunker and Nolte

Year	Uncovered Reservoir		Covered Reservoir	
	Max. Amount of Organisms found, Standard Units per cc.	Per Cent of Samples Con- taining no Organisms	Max. Amount of Organisms found, Standard Units per cc.	Per Cent of Samples Con- taining no Organisms
1916	124	3.	24	47
1917	24	91	4	85
1918	39	50	3	95

reservoirs are strikingly shown in comparative results obtained at Washington, D. C. In this city Brightwood Reservoir and Reno Reservoir both store filtered water which flows to them from the Washington Filtration Plant. The first mentioned reservoir is open, the second covered. The microscopic and bacteriological counts and determinations of albuminoid and nitrate nitrogen in the water of the two reservoirs are recorded in Figs. 99 and 100 and in Table 92. The data for the figures and the table were kindly furnished by Mr. C. J. Lauter, Chief Chemist of the Filtration Works.

Considering the microscopic counts (Fig. 99), it is seen that *Synedra* and other plankton developed often in large numbers in the open reservoir while the covered reservoir showed the presence of organisms on but two occasions and then in negligible quantities. The correlations between microscopic life and albuminoid ammonia and nitrates (Table 92), the former direct, the latter inverse, are evident. A study of the bacteriological results (Fig. 100) shows the direct effect of keeping the water away from dust and other contaminating substances as well as the indirect effect of preventing the growth and decay of plankton with its concomitant rise in bacteria.

Various attempts have been made to prevent the access of light to reservoirs by constructing cheap roofs or floating rafts of boards. It is said that in some cases they have effectually prevented the growth of *algæ*. They do not appear to have been permanently successful and their economy is questionable, except for very small reservoirs. If used at all, the entrance of sunlight through cracks between boards should be prevented.

When it is impractical to store filtered waters or ground waters in covered reservoirs, the control of algæ can be aided by designing the open reservoir in such a way that the water circulates rapidly and continuously from inlet to outlet. There should be no dead pockets. The

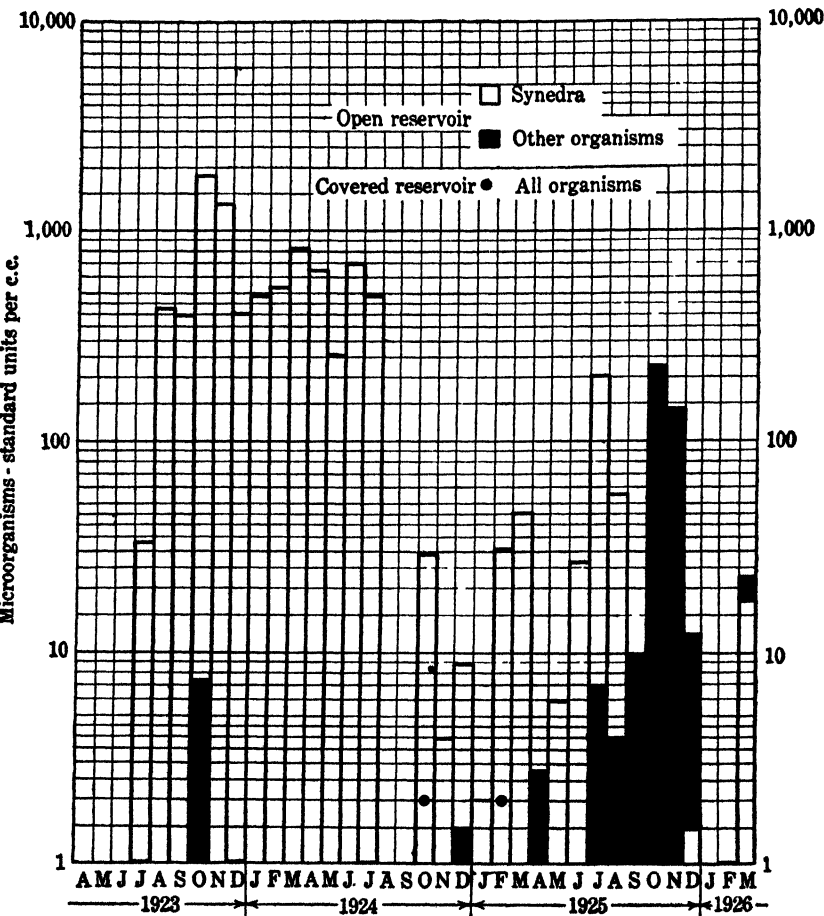


FIG. 99. — Microscopic Organisms in Open and in Covered Reservoirs, Washington, D. C. Average of Weekly Counts.

reservoir floor and walls should be smooth to prevent the growth of attached algæ and higher aquatic plants. To permit cleaning, the basin should be divided into two parts that can be operated independently. Single basins should be provided with by-pass facilities. Small reservoirs in which untreated surface waters are stored can also be designed to good advantage according to these principles.

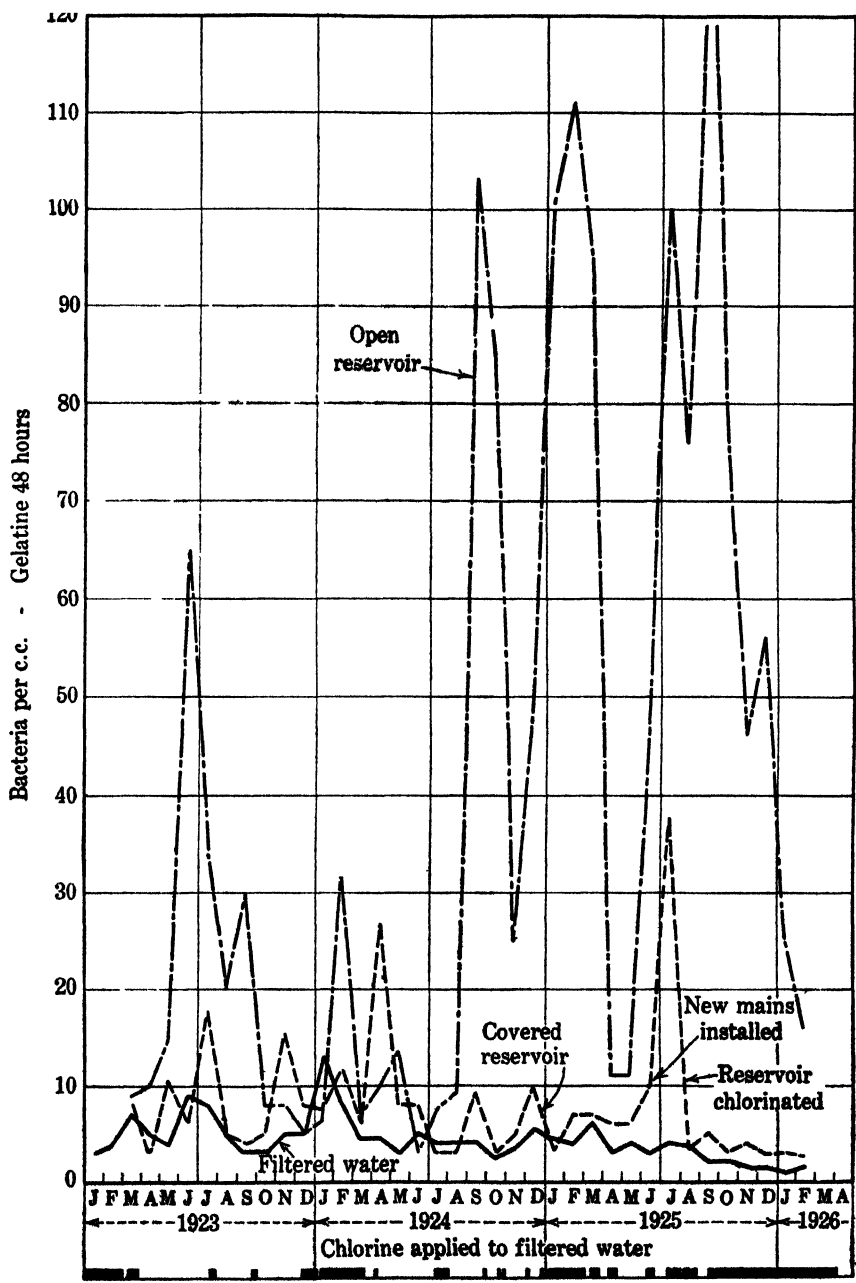


FIG. 100. — Bacteria in Open and in Covered Reservoirs, Washington, D. C., 1923-1926. Average of Weekly Counts.

TABLE 92
NITROGEN IN OPEN AND COVERED RESERVOIRS
Washington, D. C., 1925

Average of Weekly Tests — Results in Parts per Million of Nitrogen

Month	Albuminoid Ammonia				Nitrates			
	Raw Water	Filtered Water	Covered Reservoir	Open Reservoir	Raw Water	Filtered Water	Covered Reservoir	Open Reservoir
Jan.061	.031	.032	.046	.62	.67	.64	.58
Feb.067	.031	.028	.033	.57	.60	.59	.55
March.074	.034	.029	[.032]*	.62	.68	.65	.63
April.071	.029	.024	.033	.58	.63	.62	.61
May.088	.033	.021	.035	.53	.59	.60	.57
June.116	.040	.030	.045	.52	.60	.59	.54
July.104	.038	.025	[.060]	.57	.61	.59	.52
Aug.103	.039	.023	.057	.50	.59	.59	.54
Sept.109	.043	.026	.049	.42	.49	.49	.43
Oct.091	.037	.021	.052	.50	.56	.55	.48
Nov.084	.042	.026	.043	.61	.65	.57	.52
Dec.048	.038	.025	.040	.61	.64	.62	.55

* Bracketed figures signify that the reservoir was out of service part of the time.

DESTRUCTION OF ALGAL GROWTHS BY USE OF ALGICIDES

We have so far confined our attention to the prevention of algal growth or troubles by suitable physical treatment of the catchment area or reservoirs. Although much can be done in this way to reduce the number of organisms developing in reservoirs or drawn into service mains, it is either impossible or impracticable to avoid occasional growths of plankton in the best of reservoirs, except covered reservoirs storing ground water or filtered water. For full control of water supplies in open reservoirs, the use of algicides cannot be avoided and is often the quickest, cheapest and most reasonable means of preventing or remedying algal troubles.

Copper Treatment for Algæ. — In 1904, Dr. George T. Moore and Mr. Karl F. Kellerman, of the Bureau of Plant Industry, U. S. Department of Agriculture, published a report stating the results of successful experiments in which they used copper sulphate for the eradication of algæ and other microscopic organisms from reservoirs. This report immediately attracted wide attention, and the use of copper sulphate as an algicide has since become standard practice.

Copper sulphate had been used as a fungicide before Moore and Kellerman proved its worth for destroying algæ. Experiments had been made by Miquel, Devaux, and others, which showed that minute doses of poisonous substances were able to destroy the unicellular microscopic organisms, but full credit belongs to Moore and Kellerman for the use of copper sulphate in the treatment of water supplies. The first practical test on a working scale was made at the water-cress beds in Ben, Va., in 1901, where a troublesome growth of *Spirogyra* was eliminated.

Effect of Copper on the Human System. — The first question that was naturally raised when copper treatment of water was advocated was its possible deleterious effect on the human system. To controvert any argument of this kind, Moore collected extensive data to show the wide distribution of copper in nature, its presence in vegetables and even in natural waters themselves, as well as the extent to which copper salts were used in medicine. Clark also showed that some natural waters in Massachusetts contained small amounts of copper. The matter of copper poisoning has been investigated more recently by Dr. F. B. Mallory, who has established the fact that copper, when taken continuously into the system in large amounts, causes a destruction of the red blood cells and a degeneration of the liver. Small quantities, however, have so far not been proved dangerous. Experience with the use of copper in many water supplies has fully demonstrated the innocuous character of the use of copper sulphate as an algicide; the amounts used are extremely small. The application of copper to drinking water is not a matter, however, that should be left to the ordinary laborer; intelligent and continual supervision are essential. The present hygienic standards for water purity of the U. S. Public Health Service have set an upper limit of 0.2 p.p.m. of copper in drinking water.

Methods of Applying Copper Sulphate. — The methods of application are simple. Ordinary commercial crystals of blue vitriol are used. These crystals are placed in a coarse bag, gunny sack, perforated bucket, or wire basket. The container is attached to a rope and drawn back and forth in the water at the stern of a rowboat. The boat should take a zig-zag course so as to triangulate the surface of the water. An out-rigger may be arranged so as to drag two or more bags at the same time, thus cutting a wider swath. With several boats quite a large reservoir can be covered in a working day. For very large reservoirs a motor launch may be used. The churning action of the propellor is a decided advantage. Parallel paths about 100 feet apart are taken first in one direction and then at right angles thereto. Dosage must be apportioned to each unit of reservoir area under due consideration of

the depth. The speed of movement must then be adjusted accordingly. Reservoirs of 30 billion gallons capacity have been treated successfully by this simple method. Care must be taken not to row or drive the boat too slowly, as too great a concentration of copper may otherwise be obtained near the bags. Fish swimming into this overdosed water may then be poisoned. The amount of copper sulphate that will dissolve in a given time will vary with the speed of the boat and the quality of the bag. At Hartford 100 pounds were dissolved from two coarse-mesh grain bags in *five minutes* while traveling at a rate of six miles per hour. By using canvas or other material of denser weave the rate of solution can be cut down to *100 pounds or less per hour*.

It is generally preferable to carry out the treatment on a day when the wind is blowing, so that circulation of the water may more readily distribute the chemical. It will often be found best to move against the wind. Advantage may be taken also of vertical convection currents. If the algæ to be killed are near the surface the application should be made early in the day when the surface water is warming and tending to become stratified; but if the algæ are well scattered through the water strata it is better to make the application toward night when the surface strata are being cooled and tend to sink. A knowledge of the currents, such as may be obtained from Chapter VII, will be an aid to judgment in this matter. Distribution of the chemical may be assisted by suspending the bags so that they will travel through the water at different depths. The solution of copper sulphate is heavier than water and tends to sink. It has been found difficult to treat frozen reservoirs as the chemical does not diffuse readily, but precipitates to the bottom near the point of application.

Several other methods of applying copper sulphate have been used to meet special local requirements. Revis reports the treatment of two reservoirs of the Cheyenne water works by scattering the chemical on the ice in mid-winter. Taking advantage of the wind one man was able to "sow" a strip 150 feet wide at a single passage. The reservoirs had a combined area of 312 acres and a capacity of over 3 billion gallons. Dosing at a rate of 3 pounds per million gallons four men were able to complete the work in five days. The sulphate went into solution before the ice broke up. Good distribution was obtained and in no section was there a pocket of concentrated copper into which fish might swim and be killed.

In shallow bays it is difficult to apply algicides in the usual manner because the bags drag along the bottom and stir up mud which immediately takes up the copper. Under these conditions it is better to use a small pump and spray with which a solution of the chemical is thrown

onto the water surface. A tree-spraying outfit can be employed to good purpose. If the nozzle is detached, distances as great as 50 feet can be reached. Professor Huff reports using this method on Vadnais Lake where one application freed the bays from organisms.

An excellent method of applying copper sulphate when the water to be dosed passes a given point at a known rate of flow is to add the copper salt by means of chemical feeding-devices similar to those used in water purification plants for the addition of coagulants and other chemicals to

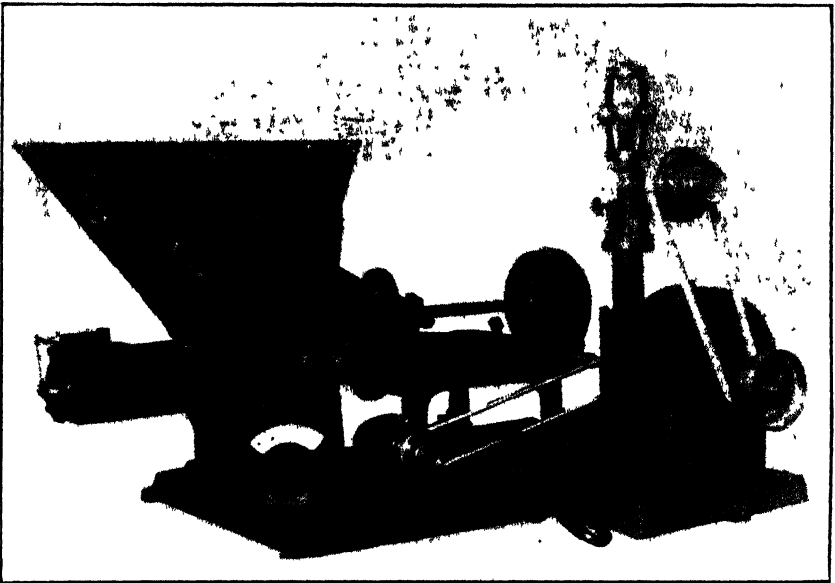


FIG. 101. — Hamblet Dry Feeding Machine. *Courtesy of Allen Hazen and Hamblet Machine Co.*

water. Dry-feed machines are particularly serviceable and a good type is illustrated in Fig. 101. This method of applying copper has been used since 1920 on the Catskill aqueduct leading to Kensico Reservoir of the New York supply. The object of dosing is to destroy organisms being fed into Kensico from the impounding reservoirs of the Catskills and thus to prevent seeding this storage reservoir near the city. Copper sulphate is applied through a dry-feed machine at Pleasantville about two hours' flow above Kensico. The dry chemical slides through a chute into a perforated wooden box floating in the water. By dosing the water whenever troubles occur in the upland reservoirs growths in Kensico Reservoir are well controlled. The period of time during which it has been necessary to apply the algicide has varied from one month

in two periods during 1921 to six months in three periods during 1923. This method becomes particularly valuable during the winter months when reservoirs cannot be dosed directly on account of being frozen over. Whenever a solution of copper sulphate is used, it is well to remember that it should not come into contact with iron since the latter will cause the precipitation of the copper.

When distributing reservoirs are to be treated with copper sulphate, they should be by-passed until the tastes and odors resulting from the death of the microorganisms have disappeared. This usually happens in about three days. In order not to lose the benefits to the distribution system afforded by the reservoirs it is customary to by-pass them by closing the influent gates only and letting the reservoir "ride" on the effluent. When no by-passes are provided the influent may be treated with copper sulphate and will then displace the untreated water. In reservoirs with five to six days' capacity, sufficient time will commonly elapse between treatment and consumption to prevent complaints from tastes and odors.

Nature of the Reaction. — Just how the copper sulphate acts in the destruction of algæ it is difficult to say, involving as it does intricate problems of cytological chemistry. The poison probably combines chemically with the cell plasm and produces compounds that interfere with the continuation of the life processes and thus cause death. The mode of action is similar to that of the so-called disinfection processes and is a function of the number of cells, the concentration of the chemical, and the time of exposure to it. As shown in Fig. 104 some cells succumb quickly while others survive a long time, the number of organisms dying in a given time interval being proportional to the number surviving.

Much interest is attached to the ultimate fate of the copper added to the water, whether or not it be involved in the reaction with the organisms. The sulphate of copper that is left free in the water reacts with calcium bicarbonate, which is present to a greater or less extent in nearly all natural waters, to form sulphate of calcium and basic copper carbonate, some carbonic acid being liberated. The basic copper carbonate may then become decomposed, copper hydrate and carbonic acid being formed. Copper hydrate is almost insoluble in water, whereas basic copper carbonate is somewhat soluble in water that contains carbonic acid, especially if the hardness is low. Experiments have shown that in hard waters the reactions above mentioned take place in the course of a few hours, the copper hydrate first becoming a colloid and then precipitating as solid matter in suspension. In softer waters more time is required. Cold and the presence of organic matter in

solution tend to retard the reaction. Heat and the presence of suspended matter tend to hasten it. The latter is probably a physical phenomenon. These are all important considerations, as the quantity of copper sulphate required to remove the algæ is closely related to the speed of the reaction.

It is difficult to state how much of the copper added to the water in reservoirs is carried into the distribution system. Some of it is precipitated but remains in suspension: some settles to the bottom of the reservoir. The proportion remaining in the water will naturally vary with local conditions. Goodnough found that the mud in the reservoir at Arlington, Massachusetts, contained as high as 0.3 per cent copper, indicating a high rate of precipitation and settling. Hale, on the other hand, concluded from a series of experiments with the New York supply that during the winter months at least all copper fed into the supply apparently came through into the distributing system. According to Hale the amounts of copper obtained, however, were too small to be of sanitary significance.

Quantity of Copper Sulphate Required. — It is of great importance that just the right quantity of copper sulphate be used. If too little is applied the algæ will not be destroyed; if too much is used, money will be wasted and there is danger of killing fish.

In deciding upon the dosage, several factors need to be considered; namely, the kind of algæ to be eradicated, the character of the water as measured by the amount of organic matter present, the hardness, the carbonic acid content and the temperature, the species of fish present, and of course the quantity of water to be treated. Some of these matters were considered in the preceding section.

It is hazardous for one not familiar with the various factors involved to attempt treating a water supply with copper. Of particular necessity is it to know what organisms are present and which of them need to be killed. For this a microscopical examination is essential. Simple equipment like that described in Chapter IV and a general knowledge of the different organisms such as may be obtained from the plates at the end of this book should in most cases be sufficient to furnish the desired information.

Quantity Required to Eradicate Different Organisms. — Organisms differ considerably in their susceptibility to copper sulphate. Some of the blue-green algæ are destroyed by the application of only one part of copper sulphate in ten million parts of water, while other organisms require more than ten and even twenty times this dose. One of the organisms most easily killed is *Uroglenopsis* which can be eradicated by using as little as 0.05 p.p.m. of copper sulphate.

It is probable that the stage of growth of the organisms is also a determining factor and that the presence or absence of carbonic acid is important. Different observers have brought in different figures for the quantities that have proved efficacious with the same organisms in different waters. It is impossible to give any very definite figures for the quantities required. The following figures which were recently assembled by Hale and which are based chiefly on values given by Kellerman, one of the originators of the method, are believed to be as reliable as any, but should be varied to meet the requirements of different waters.

TABLE 93
COPPER SULPHATE REQUIRED FOR ERADICATION OF DIFFERENT ORGANISMS

Organisms	Parts per Million	Pounds per Million Gallons**	Organisms	Parts per Million	Pounds per Million Gallons**
<i>Cyanophyceæ:</i>			<i>Diatomaceæ:</i>		
Anabaenat†§.....	0.12*	1.0	Asterionella†§.....	0.12*-0.20*	1.0-1.7
Aphanizomenon†§.....	0.12*-0.50*	1.0-4.2	Fragilaria†.....	0.25	2.1
Clathrocystis§.....	0.12*-0.25*	1.0-2.1	Melosira††.....	0.33	2.8
Cœlosphaerium†§.....	0.20*-0.33	1.7-2.8	Navicula†.....	0.07	0.6
Microcystis.....	0.20	1.7	Synedra†§.....	0.50*	4.2
Oscillatoria†.....	0.20-0.50*	1.7-4.2	Stephanodiscus.....	0.33	2.8
			Tabellaria†§.....	0.12*-0.50*	1.0-4.2
<i>Chlorophyceæ:</i>			<i>Schizomycetes:</i>		
Ankistrodesmus.....	1.00	8.3	Beggiatoa†.....	5.00	41.5
Chara†.....	0.10-0.50	0.8-4.2	Crenothrix†.....	0.33	2.8
Cladophora†.....	0.50	4.2	Sphaerotilus dichotomus.....	0.20	1.7
Closterium.....	0.17	1.4			
Cœlastrum†.....	0.05-0.33	0.4-2.8	<i>Fungi:</i>		
Desmidiæ.....	2.00	16.6	Leptomitæ.....	0.40	3.3
Draparnaldia.....	0.33	2.8			
Eudorina§.....	10.00	83.0	<i>Protozoa:</i>		
Enteromorpha.....	0.50	4.2	Ceratium§.....	0.33	2.8
Hydrodictyon.....	0.10	0.8	Chlamydomonas.....	0.50	4.2
Microspora.....	0.40	3.3	Cryptomonas§.....	0.50	4.2
Palmella.....	2.00	16.6	Dinobryon§.....	0.25*	2.1
Pandorina§.....	10.00	83.0	Euglena.....	0.50	4.2
Scenedesmus.....	1.00*	8.3	Glenodinium§.....	0.50*	4.2
Spirogyra†.....	0.12	1.0	Mallomonas§.....	0.50	4.2
Staurostrum.....	1.50	12.5	Peridinium§.....	0.50*-2.00	4.2-16.6
Tribonema†.....	0.25	2.1	Synura†§.....	0.12*-0.25*	1.0-2.1
Ulothrix.....	0.20*	1.7	Uroglenopsis†§.....	0.05*-0.20*	0.4-1.6
Volvox§.....	0.25	2.1			
Zygnema.....	0.50	4.2			

* Dosage successful in New York City's supplies.

§ These organisms have caused trouble because of odor.

† These organisms have caused trouble other than odor, such as turbidity and scum.

† These organisms have been affected by chlorine and in some cases controlled by dosages ranging from 0.5 to 2 p.p.m. depending largely on amounts of manure.

** Conversion factor: — One part per million equals 8½ pounds per million gallons.

The figures of Table 93 may be assumed to apply at a temperature of 15° C. or 59° F. If the water is colder more copper sulphate is required to do the same work, if warmer less. Moore and Kellerman state that the values given should be increased or decreased by about 2.5 per cent for each degree below or above 15° C. They also state, though with less assurance, that the values should be raised by 2 per cent for each 10 p.p.m. of organic matter and by 0.5 to 5 per cent for each 10 p.p.m. of alkalinity. A 5 per cent increase should be made if the amount of carbonic acid is small. In the past these corrections have not been applied as much as they should be to test the limits by actual experience. Better information on this phase of the problem is much needed.

When Should Reservoirs be Dosed? - Too often the dosing of reservoirs is delayed until serious troubles are experienced from the growth of microscopic organisms. This is poor practice. In a well-operated water supply system the reservoirs should be examined with sufficient regularity to permit dosing at a time when limnologic and microscopic evidence sound the warning that unless controlled there is danger of growths developing in sufficient abundance to cause trouble. *The value of analyses to regulate rather than to record performance should be more fully appreciated.*

In New York City a splendid system of control has been in operation for many years. Odor-producing organisms with the exception of *Synura* are wiped out whenever they reach a concentration of 1000 standard units. *Synura* is destroyed as soon as it is detected, no matter how small the number of organisms. Other organisms are not permitted to develop in sufficient quantities to cause turbidity, or give rise to other complaints.

When water supplies are filtered it pays to control the growth of plankton very carefully. Filter runs reflect the presence of microscopic organisms very quickly and the cost of more frequent scraping in slow-sand filters and greater use of wash water in rapid-sand units can often be offset by more rigid reservoir control (see Chapter XIV). Limnologic studies are a valuable aid to control by heralding the overturns, locating the circulation, transition and stagnation zones, and supplying other important data in regard to dosing requirements.

Calculating the Volume of Water to be Treated. - The quantity of water to be treated is readily computed from a contour map of the reservoir or lake bed such as the one shown in Fig. 102 for Fresh Pond, Cambridge. The average end area method* of calculating volumes is sufficiently accurate for calculating the amount of water in the reservoir, and a capacity curve (Fig. 103) can be drawn in order to permit the

* See any standard treatise on surveying.

rapid determination of quantities up to and between different reservoir levels. It may be well to divide the reservoir surface into several areas of approximately equal depth in order to adjust to best advantage the dose and rate of application. This division becomes necessary whenever shallow bays or reservoir arms are to be dosed independently of the main body of water. Separate calculation of volumes and separate capacity curves are then required.

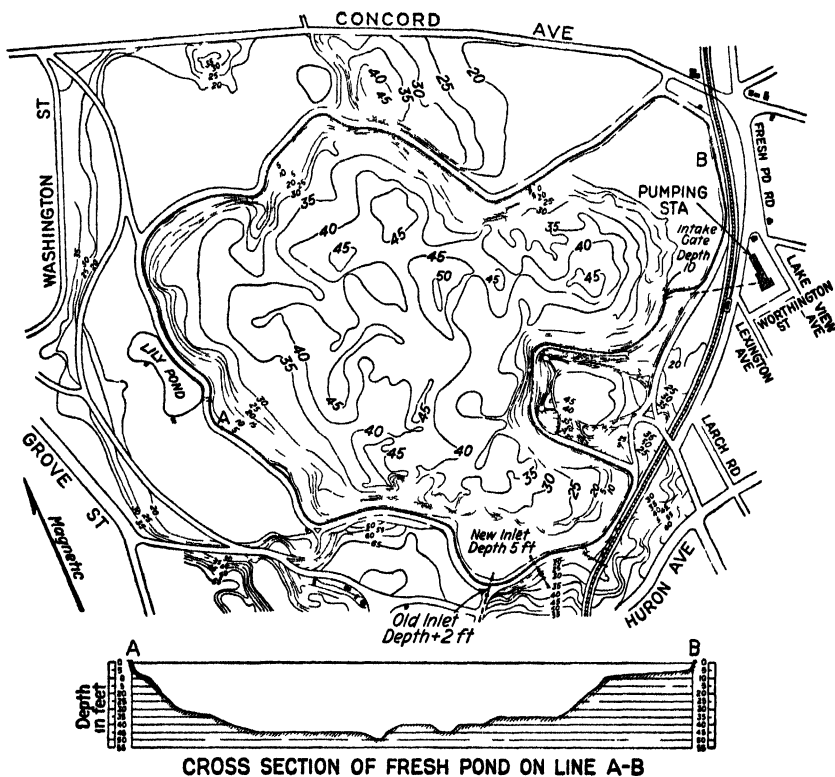


FIG. 102 — Map of Fresh Pond Reservoir, Cambridge, Mass.

If the reservoir to be treated is so deep that the lower strata are stagnant, only the water above and within the transition zone should be included in the calculations. This involves a knowledge of the temperatures at different depths which may be obtained by the methods described in Chapter VII.

When hydrographic maps of the lake or reservoir are not available the volume must be estimated as closely as possible. The following rules will assist in making the necessary approximations.

One million gallons of water represent a depth of about 3 ft. over one acre. Hence the number of acres of water surface, multiplied by one-third the average depth of water in feet gives approximately the number of million gallons of water in the reservoir. In most reservoirs the average depth may be taken as about one-third the maximum depth.

When the volume of water expressed in million gallons has been found, the poundage of copper sulphate required is ascertained by multiplying

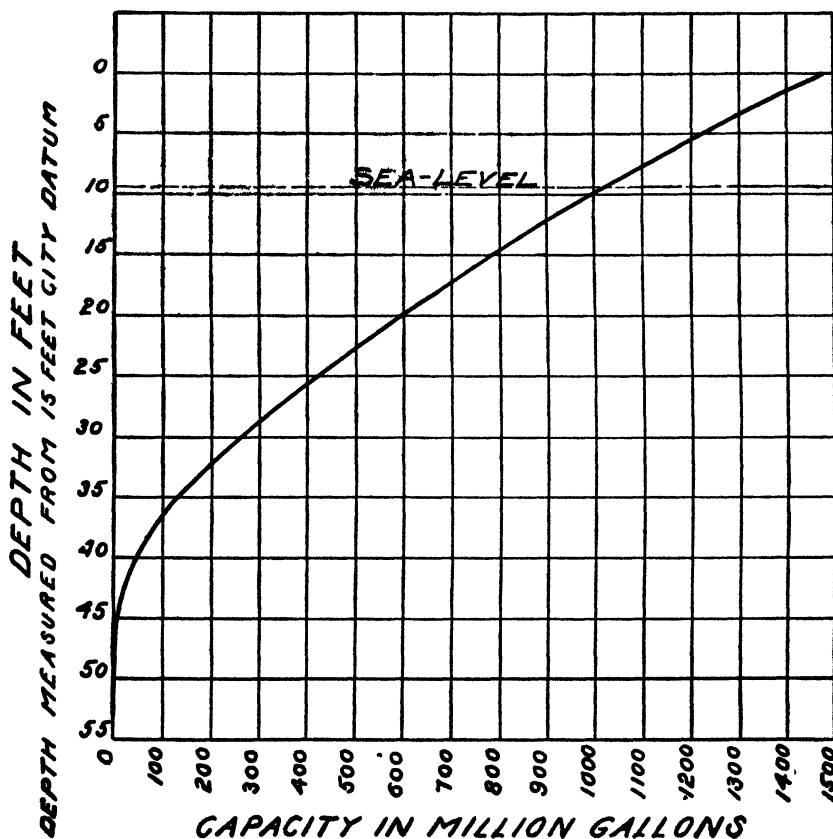


FIG. 103. — Capacity Curve of Fresh Pond Reservoir, Cambridge, Mass.

the water volume by that value in the last column of Table 93 which applies to the organism to be killed. This quantity must then be corrected to take account of temperature, organic matter, alkalinity, and carbon dioxide.

Death of Fish in Copper-Treated Water. — Kellerman has studied the effect of copper sulphate upon fish and recommends the following limits of treatment in the presence of certain kinds of fish (see Table 94).

TABLE 94
CONCENTRATION OF COPPER SULPHATE THAT WILL KILL FISH

Fish	Parts per Million	Pounds per Million Gallons (Approx.)
Trout.....	0.14	1.2
Carp.....	0.30	2.5
Suckers.....	0.30	2.5
Catfish.....	0.40	3.5
Pickarel.....	0.40	3.5
Goldfish.....	0.50	4.0
Perch.....	0.75	6.0
Sunfish.....	1.20	10.0
Black bass.....	2.10	17.0

It will be seen that some of the amounts required for the destruction of microscopic organisms are close to the doses that will kill fish. Even when relatively small quantities of the chemical are added to reservoirs, however, there is danger of destroying those fish which by chance swim into the relatively concentrated solution of copper sulphate that issues from the bags as they are drawn through the water. The number killed, in this way, however, is usually small.

When very large numbers of plankton are destroyed by the application of copper sulphate the dead organisms are apt to accumulate upon the gills of fish and to smother them. Thus Hale noted the death of fish in quantity upon the treatment of 20,000 units of *Synedra*. Examination of the fish, chiefly perch, showed that their hearts were still beating while their gills were covered with the dead growth to such an extent as to prevent breathing. Death due to smothering usually occurs on the second or third day following treatment. The depletion of oxygen dissolved in the water following the destruction and decay of plankton may here be a contributing factor (see Chapter VIII).

When well supervised the use of copper sulphate should not give much trouble by destroying fish life. In the treatment of the water supplies of New York City, fish have only rarely been killed in any quantity and the dosage has varied from 0.05 to 0.5 p.p.m. On rare occasions it has been even higher.

After-Growths of Organisms. — It happens not infrequently that after copper sulphate has been used to destroy a certain organism a new growth of some other kind appears. One type of plankton may be succeeded by another, and bacteria commonly flourish after algæ are destroyed.

After-growths of Plankton. — After-growths of plankton are exemplified by the increase in diatoms that frequently follows the destruction of blue-greens such as *Anabæna*. Usually the second growth is an organism less susceptible to the influence of copper than the first, but sometimes the same species returns. There is no evidence, however, that organisms acquire resistance to copper sulphate and that doses increasing with each treatment are needed to kill them.

The development of after-growths of plankton is illustrated by the following figures taken from the records of the Cambridge, Massachusetts, Water Works. On July 14, 1925, Fresh Pond contained 156 standard units of blue-green algæ. A dose of copper sulphate applied on July 18 resulted in their reduction to 128, 116 and 32 units after 1, 4 and 6 days, respectively. On the seventh day an after-growth of diatoms appeared measuring 132 units. This growth increased to 290 units after 5 days and to 748 units, largely *Synedra*, after 19 days. A similar experience was recorded in the preceding year, when blue-greens measuring 1000 standard units were dosed on August 4 and decreased to 700 and 468 units in 1 and 4 days, respectively. *Synedra* appeared 10 days after dosing and increased to 2680 units 3 weeks after the chemical was added. The pond was then given another treatment which reduced *Synedra* to 508 units.

In treating reservoirs it is well to remember that organisms sometimes become concentrated within the transition zone, whence they may be carried into the circulating waters by high winds and cooler weather. Such growths should be watched so that subsequent treatment may be given as soon as the organisms begin to multiply.

Increase of Bacteria after Copper Treatment. — A secondary effect of copper treatment is to increase the number of bacteria in the water. The following figures by Jackson illustrate this bacterial rise. They refer to one of the reservoirs of the water supply of Brooklyn that had been treated with copper to destroy a growth of *Asterionella*.

An analysis of these figures yields some interesting information, particularly when the values are plotted on *semi-logarithmic* or *ratio* paper as has been done in Fig. 104. If we confine our attention first to the effect of copper sulphate treatment on the plankton it becomes apparent that the use of the algicide does not result in the instantaneous death of all the organisms. Some of the cells survive for quite a long time, and it stands to reason that the gradual removal of the poison from the water by its action on the organisms, combined with its precipitation as a result of chemical reactions with other substances in the water, will permit some of the plankton organisms to escape destruction. This, together with the fact that apparently the most resistant organisms

survive, explains why reservoirs are seldom permanently rid of algal growths. The rate at which the organisms die is strikingly uniform, as evidenced by the fact that the values plot so close to a straight line on ratio paper. This is quite in accord with the laws governing disinfection

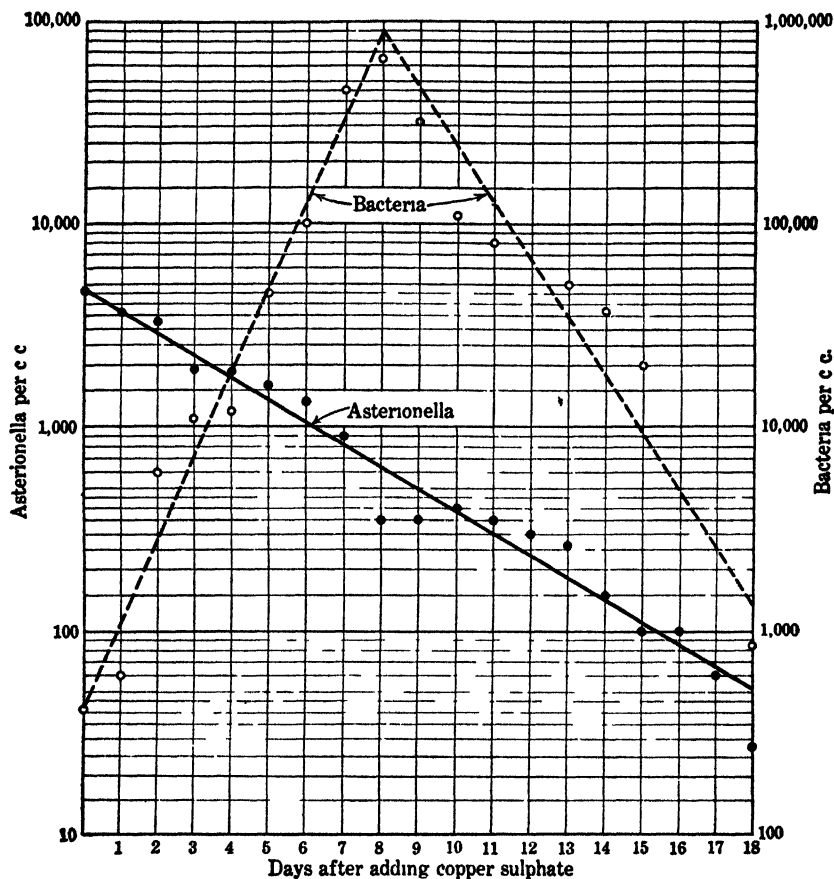


FIG 104 — Effect of Destruction of *Asterionella* on the Numbers of Bacteria.

tion and there is more than a mathematical reason why the curve representing the death of the organisms should be called the "die-away" curve. With the dose of copper sulphate used in this case about 20 per cent of the surviving organisms succumbed during each day. The destruction of microscopic organisms by copper sulphate, like all "disinfection" processes, is due to a time-concentration effect.

As soon as the plankton organisms die the carcasses furnish food for saprophytic bacteria which begin to multiply at a very rapid rate. This increase is further aided by the fact that many food materials used

TABLE 95
EFFECT OF THE DEATH OF ASTERIONELLA ON WATER BACTERIA

Date		Number per Cubic Centimeter	
		Microscopic Organisms	Bacteria (20° C.)
March 13, 1905	Before treatment	4625	405
14	After "	3645	600
15	" "	3325	6,000
16	" "	1925	11,000
17	" "	1850	12,000
18	" "	1575	45,000
20	" "	1350	100,000
21	" "	900	440,000
22	" "	350	630,000
23	" "	350	310,000
24	" "	400	107,000
25	" "	360	80,000
26	" "	300	64,000
27	" "	270	50,000
28	" "	150	37,000
29	" "	100	20,000
30	" "	100	8,000
31	" "	60	3,500
April 1	" "	28	860

jointly by algæ and bacteria become available to the latter alone. A point is finally reached when the water cannot support the tremendous bacterial flora and the bacteria are then gradually reduced in number. It is, of course, the normal water bacteria, as determined by the 20° C. count, that show the most striking variation. As shown in Fig. 95 their development follows the law of organic growth analogous to compound interest; their subsequent death similarly manifests the characteristics of organic death; i.e., it follows the die-away curve.

Sometimes the numbers of bacteria are even higher than those given in Table 95. Bacterial growth following the use of copper may be reduced by dosing the water with hypochlorite of lime or any other chlorine compound. Copper sulphate in sufficient concentration will itself destroy bacteria. The amount required, however, is considerably greater than that needed to destroy algæ. For killing bacteria copper sulphate is less efficient than are chlorine compounds.

Subsequent Odors of Decomposition. — The decay of the algæ after they have been killed causes a temporary increase in the odor of the water. When the correct dosage of copper sulphate has been applied the odor is only of short duration, and the reservoir is usually ready to be turned into service on the third day after treatment. Best practice requires that laboratory tests of water quality be made before the water is allowed to pass to the consumers. Complaints due to unforeseen conditions can thus be avoided.

Examples of Reservoir Control by Copper Treatment. — A discussion of the use of copper sulphate as an algicide would not be complete without the citation of a few examples of observations covering the behavior of water supply reservoirs before, during, and after control. While each catchment area and reservoir is more or less a law unto itself, much can be learnt by the analysis of records from different sections of the country. From these records or reports, methods of procedure can be checked against results obtained; opinions as to the behavior of different kinds of organisms can be formulated; relative values of treatment can be established. Familiarity with the experience of others develops good judgment, and knowledge of the practice of others stimulates the critical faculties.

St. Paul Experience. — In 1915, Professor N. L. Huff was employed by the city of St. Paul to study the microorganisms in Vadnais Lake and to treat the water, if necessary, with copper sulphate. The first treatment was applied on June 14 (Fig. 105). The total count was 8100 standard units per cc., 3420 units being *Synedra pulchella*. A dose of 0.08 p.p.m. CuSO_4 was applied by launch. In less than two weeks the count dropped to 2400 standard units with 1100 *Synedra*. Early in July, *Synedra* began to increase and by July 12 had reached 8260 units in a total count of 9700. The water was then dosed with 0.1 p.p.m. CuSO_4 which in ten days reduced the count to 100. Another increase in August resulted in the development of 4000 units, the addition on August 27 of 0.1 p.p.m. of copper again bringing the count down to 100, this time in 18 days. In October the fall overturn occurred and with it diatoms began to flourish. *Stephanodiscus* reached a maximum of 11,850 units in November but dropped to 4920 by December 11.

Diatoms were greatly affected by the first treatment, *Cyclotella* being completely destroyed. *Melosira* was reduced by the first dosage and apparently wiped out by the second; so were *Asterionella* and *Fragilaria*. The two latter, however, indicated a resumption of activity after the second treatment but were eliminated by the third.

On June 14, 500 standard units of *Spirogyra* were recorded. They were eliminated in four days by the first dose and reached a count of 150 before the second, which completely destroyed them. *Eudorina* and *Pandorina* were less sensitive, decreased after the second application and were affected but slightly by the third. Two weeks after the third

dose they were present in concentrations of 1200 and 700 units, respectively, and then suddenly dropped to 100 units.

The blue-greens were greatly affected by the first application and were eliminated by the second. *Anabæna* was especially sensitive and frequent application of copper sulphate in sheltered bays kept these forms in check and prevented their spreading to other parts of the lake.

Of the protozoa, *Ceratium*, *Dinobryon*, *Vorticella*, and *Uroglenopsis* disappeared 10 days after the first treatment. Reappearing in small numbers, they were wiped out by the second dose.

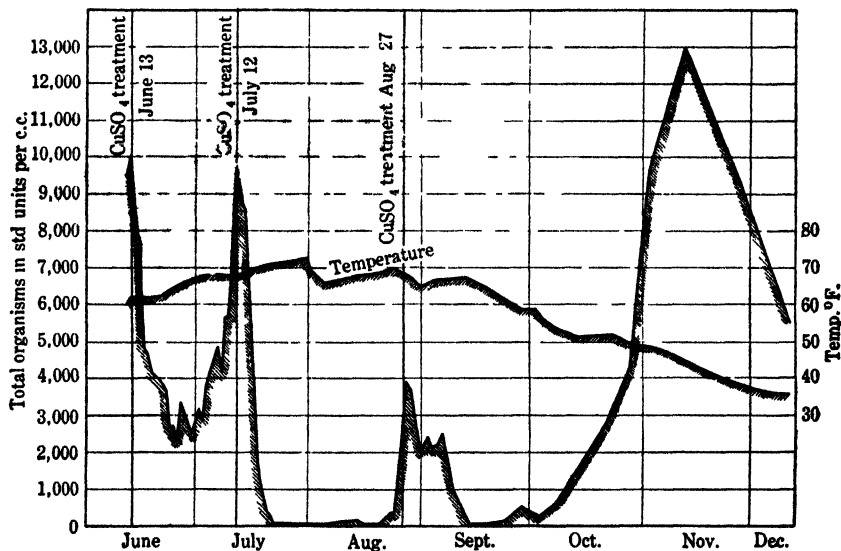


FIG. 105 — Results of Copper Sulphate Treatment of Vadnais Lake, St. Paul, Minn., 1915. After Huff.

Figure 105 shows graphically the history of the organisms during and after treatment.

Rockport Experience.—E. Sherman Chase reports the successful treatment of Cape Pond from which the water supply of Rockport, Massachusetts, is taken.

On May 22, 1922, 0.28 p.p.m. of copper sulphate was applied to this pond. No reduction of organisms taking place, this was followed by 0.60 p.p.m. five days later. By June 8 a marked decrease had taken place, but the relatively high cumulative dose also killed a large number of white perch. Growths remained suppressed until the fall overturn when diatoms reached a concentration of 11,000 per cc. A dose of 0.5 p.p.m. of copper sulphate on November 3 reduced the count to 17 by December 26. In 1923, organisms did not become abundant until August. The addition on September 4 of 0.71 p.p.m. CuSO_4 decreased the concentration of organisms from 1000 to 168 per cc.

As shown graphically in Fig. 106, the treatment of May, 1922, reduced the growths below the previous year's record until the fall over-turn resulted in a most prolific growth of diatoms. In the following year, however, the treatment of the preceding winter and the following summer suppressed all growths of any magnitude.

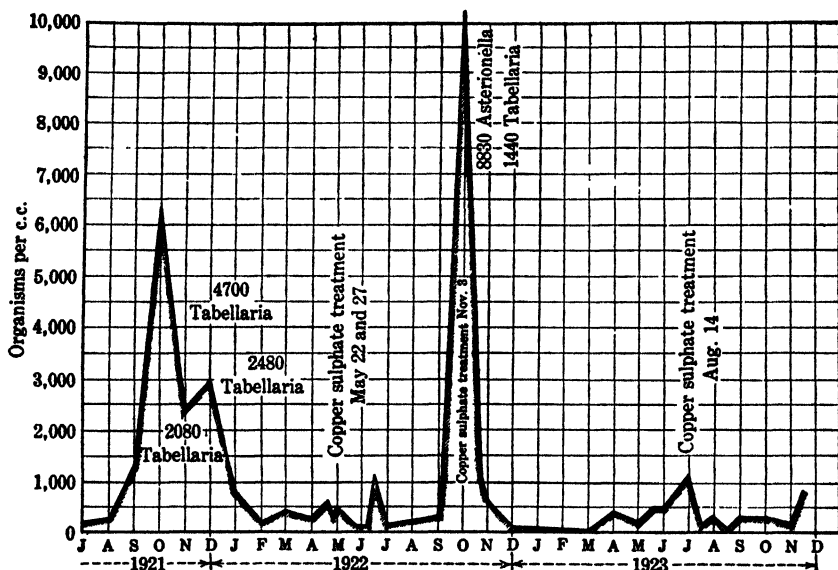


FIG. 106. — Results of Copper Sulphate Treatment of Cape Pond, Rockport, Mass., 1921-1923. *After Chase.*

Growths on Reservoir Walls. — Some of the attached algæ and higher aquatic plants have at times given trouble by growing on the walls of masonry reservoirs. The higher rooted plants naturally grow only when it is possible for their roots to penetrate into soil between the stone paving of the reservoir, but the filamentous algæ find sufficient hold even on concrete slopes. The former can only be destroyed by copper sulphate that is sufficiently strong to penetrate to the roots of the plants. At Richmond, Indiana, it was necessary to apply 100 pounds of copper sulphate to the masonry in order to destroy a growth of *Chara*. The walls were later lined with concrete and no trouble was experienced thereafter.

Attached algæ can be eliminated by dosing the water or better by applying strong solutions of copper sulphate to the walls. At Kansas City a growth of *Spirogyra* was at one time destroyed by the use of 1.5 p.p.m. of copper sulphate in the water. Later it was found that spraying a 5 per cent solution of copper sulphate onto the walls after

the reservoir had been drawn down below the algal line yielded better results. A paint spray was used for this purpose and bi-monthly treatment kept the walls entirely free from attached algæ.

Chlorine Treatment for Algæ. — Chlorine, today the most useful water disinfectant, is also a good algicide. Its value as such was first noted by Houston during the treatment of the Lincoln, England, supply in 1905. Since then it has been studied by many different observers. The most notable work in the field of chlorination, however, has continued to be done by Houston in the laboratories, reservoirs, purification works and distribution system of the London Metropolitan Water Works. The reader will do well to acquaint himself with these studies which have appeared in the Annual and Research Reports of the London Metropolitan Water Works. In America the full use of chlorine for the treatment of algæ-laden waters was first developed in 1921 by Hale in connection with the elimination of tastes and odors from the New York water supply. The chlorination of water introduces so many questions of odor and taste elimination that are akin to those encountered in plankton eradication that a somewhat broader statement of the problem becomes necessary in the following paragraphs.

Methods of Applying Chlorine. — Whereas copper sulphate is more commonly applied directly to the water in reservoirs, chlorine as an algicide has to date only been added to the water as it passes certain control points. This does not mean, however, that it is impossible to dose storage reservoirs with chlorine. Numerous methods of doing so suggest themselves. If chloride of lime is used, it can be dragged through the water in bags, or it can be dissolved and sprayed upon the water surface. If liquid chlorine is to be applied the chloro-boat of Wallace and Tiernan or a similar device might well be employed.

The principal sources of chlorine are three in number as follows:

1. *Liquid chlorine* or *chlorine gas* compressed in steel cylinders and applied to water by numerous devices, one of which is shown in Fig. 107.
2. *Bleaching powder* or *calcium hypochlorite*, a solid compound with about 30 per cent available chlorine when dissolved.
3. *Electrolytic chlorine* obtained *in situ* from the electrolytic decomposition of common salt (NaCl) as chlorine gas or as liquid sodium hypochlorite.

Of these, liquid chlorine is today most commonly employed. The methods of using liquid chlorine, applying bleach and producing electrolytic chlorine will not be touched upon here, since most water supply texts give full and detailed discussions of theory and practice of water chlorination.

Nature of the Reaction. — The action of chlorine upon algæ is probably similar to that of copper sulphate, i.e., the chemical has a specific toxic effect and causes the death and disintegration of the organisms.

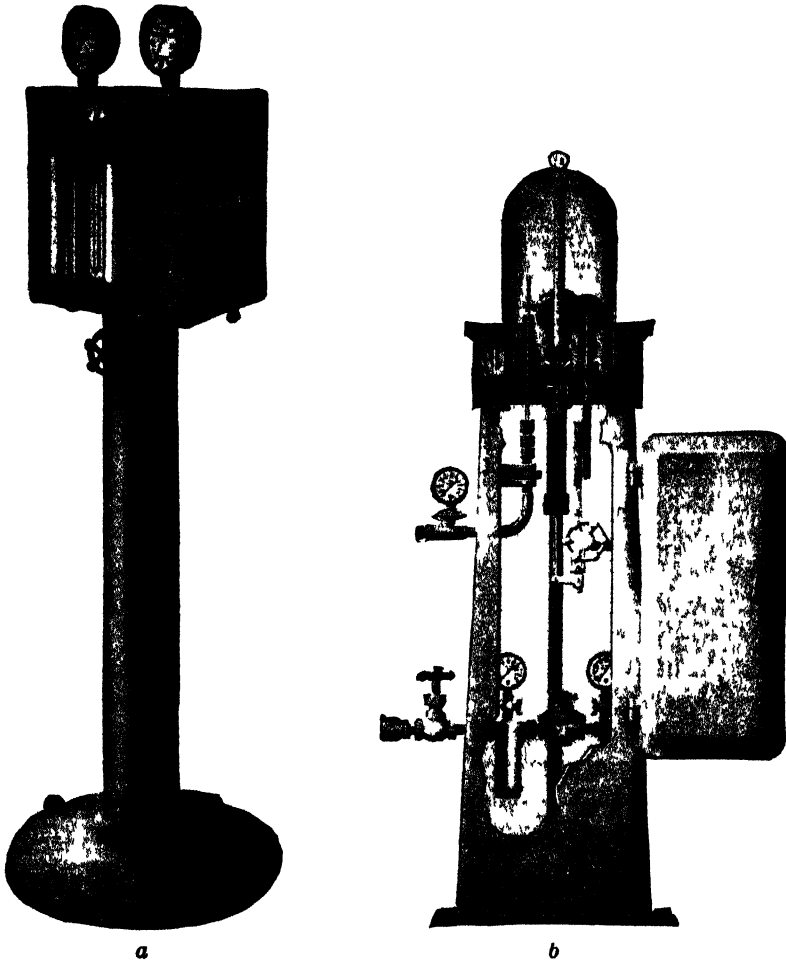


FIG. 107. — Chlorinator.

- a.* Wallace and Tiernan Solution Feed Chlorinator for the chlorination of water supplies up to 2 million gallons per day.
- b.* Wallace and Tiernan Vacuum Type Solution Feed Chlorinator for the chlorination of large water supplies

The essential oils are thus liberated and under certain conditions remain free in the water to cause odors and tastes in proportion to their concentration. In other cases, they, together with the organic matter of

the dead algæ, combine with the chlorine to form new or intensified aromatic tastes. It is this intensification of the odor and taste that makes the control of algæ in water supplies that are disinfected with chlorine — and most of our surface supplies are thus disinfected — a most interesting problem which challenges the ingenuity of the controlling laboratory.

Superchlorination. — When chlorine is added to water in excess of the quantity required to satisfy the chlorine demand of the water and to kill the microscopic organisms, the odor and taste producing oils are destroyed at the same time. Whether the excess chlorine oxidizes the aromatic substances or reacts with them to form odorless compounds is not known. The method is called superchlorination and presents a relatively new solution of the algal problem. In America it was first used with great success at New York to destroy the odors and tastes produced by the growth of *Synura* in the Catskill Water Supply.

Superchlorination is equally successful in coping with iodoform tastes. Here, too, an excess of chlorine avoids the production of the taste and will destroy any taste produced by small doses of the chemical. Chlorine therefore is in itself a cure for this group of chlorine troubles. There seems to be no upper limit to the quantity of excess chlorine that can be added without reaching a range of new odor and taste phenomena. It stands to reason, however, that too great a dose will be wasteful and may result in the formation of indeterminate odors by combining with organic materials in the water.

The chlorinous taste left in the water by the excess use of chlorine must commonly be removed by dechlorination, but, if enough time elapses before the water is consumed and the chlorine demand of the water is sufficient, the excess will pass away of itself. As in copper sulphate treatment the destruction of microscopic organisms by chlorine is a time-concentration phenomenon.

Dechlorination. — The removal of excess chlorine is known as dechlorination. There are many “anti-chlors” that can be used. The cheapest is probably sulphur dioxide which can be generated from sulphur *in situ* or purchased similar to chlorine gas in metallic containers. Other anti-chlors are sodium sulphite, bisulphite and thiosulphate. An excess of the anti-chlor can be used with safety as of itself it confers no taste on the water.

Use of Potassium Permanganate. — During his extensive researches into chlorination tastes and odors Sir Alexander Houston found that iodoform tastes can be prevented by the addition of minute doses of potassium permanganate. The dose required varies with the oxidizability of the particular water involved. In some waters 0.2 p.p.m. is

sufficient, in others much larger quantities are needed. The chemical usually imparts to the water a faint red tinge which in the presence of sufficient oxidizable material disappears as the reaction is completed. In ground waters the pink color may persist, and with colored waters a lasting brown stain may be formed. Permanganate does not retard the action of chlorination but assists sterilization. Like chlorine in excess permanganate is both a taste preventer and a taste remover. It can be added to the water before chlorination to prevent taste production or after chlorination to remove tastes produced. The point of application of the chemical can, therefore, be so arranged as to suit best existing conditions. When the water is filtered, however, one combination of dosage must be avoided, namely, permanganate treatment prior to filtration followed by chlorination. This, because of the fact that the permanganate is either used up in oxidizing the organic matter in the raw water or is absorbed in the filter so that the organic compounds (including phenols), leached out of the sand after the efficacy of the permanganate has been destroyed, combine with the chlorine to form obnoxious tastes. The observation that tastes sometimes occur on the periphery of distribution systems, i.e., after the permanganate has been dissipated, substantiates this theory.

Permanganate is not an anti-chlor; on the contrary it renders chlorinous tastes more pronounced and has *per se* an objectionable taste. Unless used with discretion it may augment taste troubles. The application of permanganate for the prevention and removal of aromatic tastes intensified by chlorination has not yet been investigated.

Use of Ammonia. — The use of ammonia compounds in connection with disinfection by chlorination was developed by Rideal and Race. More recent laboratory experiments show that, besides aiding disinfection, liquid ammonia and ammonium chloride or sulphate seem to be taste preventers. Concentrations of less than 0.2 p.p.m. are apparently effective in eliminating iodoform tastes. Their action may be similar to that of vegetable matter which, it has been shown, seems to constitute an anti-taste body. Ammonia compounds are probably of greatest value in the treatment of water, such as well water, that contains little oxidizable matter. A "lagging" effect on disinfection has been noted, and a greater period of contact between the water treated with ammonia and chlorine must be provided in order that the water may not reach the consumer before sterilization has been completed.

Quantity of Chlorine Required to Eradicate Plankton. — As in the case of copper sulphate, the resistance of microscopic organisms to chlorination is specific. Different organisms succumb to different concentrations of the chemical and there is appreciable variation in the

viability of members of the same species. This variation can probably be traced to their stage of development and other structural influences. The relative number of microscopic organisms in the water is probably of moment in determining the necessary chlorine dose.

Data on the use of chlorine are extremely meager as shown in Table 96, in which are brought together those results given in the literature which are sufficiently specific for record. (See also Table 93.) Practically all of the figures are based upon Hale's valuable work.

TABLE 96
AMOUNT OF CHLORINE REQUIRED TO DESTROY MICROSCOPIC ORGANISMS

Organisms	Concentration of Organisms. Standard Units	Chlorine Dose		Reduction in Organisms. Per Cent	Odors and Tastes Eliminated
		p.p.m.	lbs. per M. gal.		
Aphanizomenon.....	1500	0.85	7.1	50
Cyclotella.....	...	1.0	8.3	100
Melosira.....	*	2.0	16.6	100
Crenothrix.....	0.54	4.5
Fungi.....	0.33	2.7
Dinobryon.....	500	0.5	4.2	100	Yes
Uroglenopsis.....	2000	0.5	4.2	100	Yes
	6000	0.5	4.2	100	Not
	50	0.3	2.5	100	Yes
Synura.....	100	0.5-0.7	4.2-5.8	100	Yes
	200	> 0.7	> 5.8	100	Yes
Gnats of Blood Worm	3.0	24.9	100

* Present in sufficient numbers to clog filter.

† Taste not noticeable after 10 to 15 miles' flow in aqueduct.

New York Experience with Chlorination. — The following summary of experience in connection with plankton tastes and odors in the Catskill water supply of New York gives some interesting information about both chlorination and copper sulphate treatment.

In August, 1921, *Synura* appeared in Ashokan Reservoir and developed to about 100 units by October. Kensico Reservoir was next seeded and about 100 units were present in its waters by the middle of December. Complaints of fishy tastes were reported from upper Manhattan, and, on being traced back to the chlorination plants, it was found that a dose of 0.3 p.p.m. was killing the organisms and liberating the oils. Two days' storage of the chlorinated water in Hillview Reservoir did not

remove the tastes, which even passed through a double charcoal and sand filter in the Municipal Building in Lower Manhattan. The freezing of Kensico Reservoir made direct dosage of the water impossible and automatic feeding of copper sulphate into the aqueduct waters passing into Kensico was resorted to. At this time 50 units of *Synura* were coming from Ashokan Reservoir. Since this was a smaller concentration than in Kensico the latter was by-passed, but copper sulphate treatment followed by chlorination continued to produce the taste of the organism. An attempt was now made to preserve the *Synura* cells from destruction so that the oils might not be set free. The Kensico aëerator was shut down and copper sulphate treatment discontinued. The chlorine dose was lowered to 0.1 p.p.m. An almost tasteless water was produced at Hillview but the destruction of the organisms in the distribution system continued to give rise to complaints. Copper sulphate treatment was then shifted to the Ashokan gate chamber whence the water had to pass through 92 miles of aqueduct before reaching Hillview Reservoir. A dose of 0.12 p.p.m. of copper sulphate killed about 60 per cent of the organisms but the water still had a cucumber taste at Hillview. The dose was then raised to 0.18 and finally to 0.25 p.p.m. when *Synura* was eradicated and the water was rendered completely tasteless by the time it reached Hillview Reservoir. Kensico aëerator was put back in operation and the chlorine dose increased to 0.3 p.p.m., which assisted in reducing the taste. No trouble was experienced after this.

Other methods of control were now investigated and it was found that 0.6 to 0.75 p.p.m. of chlorine applied at Kensico eliminated the cucumber taste. Superchlorination was successful and the chlorinous taste disappeared in 12 to 24 hours. On March 30 and 31, Kensico Reservoir was treated with 6000 pounds of copper sulphate and turned into service on the following day. No tastes resulted.

Relative Values of Chlorination and Copper Treatment. — The successful use of chlorine as an algicide naturally leads to an inquiry into the relative merits of chlorination and copper treatment for the control of algæ. At the present time it is neither possible nor necessary to lay down fundamental rules governing the use of these two chemicals. Much work must yet be done and wider experience accumulated before definite procedures covering all possible circumstances can be established. Those in charge of reservoir operation should cultivate a spirit of research which will not only aid them personally in determining the best method of algæ control for their own needs but will also lead to new discoveries of practical value to others.

It appears that copper sulphate and chlorine will yield good results alone or in combination. The method of treatment adopted, therefore, must depend upon the facilities for dosing the water with chemicals and the general arrangement of the reservoir system. Finally, cost of operation must be balanced against ease of manipulation and results.

The use of chlorine in sewage treatment for the control of algal and other slimes on sprinkling filters and for the destruction of *Psychoda* has recently been developed.

Lime Treatment for Algæ. — The addition to water of excess hydrate alkalinity also brings about the destruction of algæ. This is probably due to the removal of the carbon dioxide (both free and half-bound) necessary for the life process of the plankton. Changes in hydrogen ion concentration resulting at the same time may also be of moment. The efficiency of this method is not well established and attempts to reduce algal growths in masonry reservoirs by drawing them down and white-washing the walls have met with varied success.

There are undoubtedly certain other chemicals that will affect algal growths. At Antwerp, for example, an open reservoir has remained consistently free from microscopic organisms. This is laid to the use on the slopes of clinker from nickel furnaces in place of the customary stone riprap.

Control of Algæ in Swimming Pools. — Algæ will develop in swimming pools as well as in reservoirs. Limnetic forms are found in the open body of pool water and attached types occur on the pool walls. In indoor pools green slimy growths often appear in places where sunlight strikes the water. In outdoor pools the whole basin is sometimes thus affected. The method of control must naturally be the same for pools as for reservoirs. Both copper sulphate and chlorine can be used although excessive chlorination is more apt to result in complaints from smarting eyes. Attached forms will frequently require draining of the pool and scrubbing of the walls with a strong copper sulphate, chlorine or lime solution. At Belmont, Massachusetts, growths of *Daphnia* and algæ in an outdoor pool have been successfully combatted with copper sulphate.

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CHAPTER XIV

PURIFICATION OF WATER CONTAINING ALGÆ

While it is possible, by suitable construction or operation of reservoirs, to control at the source the growth of algæ in water supply systems, it is often more convenient and economical, and sometimes necessary, to supplement algæ control by purification of the water collected or to adopt purification processes in lieu of certain features of reservoir construction or other measures of control. In general, purification and control do not constitute separate approaches to the solution of the problem of algæ in water supplies. Each has a close bearing upon the other and the selection of control methods should be guided by the purification processes to be used, while in turn the choice of purification works must be varied to suit the conditions of control. The removal of algæ, furthermore, is seldom the only factor to be considered in obtaining a clean water supply. More often there are other elements, such as pollution, color, turbidity, or mineralization, to which the presence of algæ is only incidental. The most satisfactory and economical solution of each water supply problem requires the striking of a balance in which each factor receives consideration weighted according to its just importance.

AËRATION

Of great success in removing odors and tastes from water is *aëration*. By this is meant the exposure of water to the air in thin films, in drops, or as a fine spray. The object is to provide opportunity for an interchange of gases and volatile substances between the water and the air. By aëration oxygen is taken into solution by the water while carbon dioxide and odoriferous gases, such as hydrogen sulphide, are released and volatile odor- and taste-producing substances are liberated.

Removal of Odors and Tastes by Aëration. — While aëration “sweeps out” the odors and tastes caused by algæ, it does not to any extent remove the organisms themselves. Even violent aëration, such as is obtained in spray aërotors, will destroy only the more fragile plankton as *Synura* and *Uroglenopsis*. New odors and tastes will be generated by the organisms that escape destruction unless they too are killed or removed. This can be accomplished by the use of algicides prior to

aération or by the construction of aërotors in connection with filtration works.

Stagnation odors and tastes obtained when water is drawn from the deeper layers of reservoirs or from streams that are covered with ice are readily removed by aération. It has been shown that deep draft is sometimes of value in securing the benefits of more desirable temperatures or in avoiding draft from water strata rich in microscopic life. The stagnation odors in the water drawn, however, may make the shifting of draft impractical unless the water can be aërated. While "sweeping out" the odors of decay, aération will furthermore cause oxidation and precipitation of the iron which is often taken into solution in the stagnant layers within which oxygen has become depleted.

Less marked is the effectiveness of aération in dealing with odors and tastes due to industrial wastes such as the phenols, cresols, or similar bodies that cause the iodoform taste in chlorinated water. These substances, apparently, are not sufficiently volatile to be removed by aération. Free chlorine, which causes chlorinous tastes, escapes more readily.

Aération as an Aid to Water Purification Processes. — Apart from the removal of odors and tastes, aération is of benefit to water purification in some other ways. Thus it is frequently a necessary adjunct to filtration or contact processes in the treatment of iron- or manganese-bearing waters. Here it provides the oxygen required to oxidize and throw out of solution these metallic substances. At the same time it removes the carbon dioxide which renders the metals soluble and makes the water aggressive. It also liberates hydrogen sulphide which is so often associated with iron-bearing waters. Sulphureted hydrogen not only imparts a foul odor to water but reacts with chlorine which may be required to disinfect the water. Removal of hydrogen sulphide, therefore, will reduce the quantity of chlorine needed.

Aération is frequently an aid to coagulation. When made to follow immediately upon the addition of alum, it is used in a number of plants to hasten floc formation and reduce the quantity of coagulant required. Here, again, it removes carbon dioxide which is freed in the reaction. For the purpose of delivering a less corrosive water to the distribution system, aération can often be employed to good advantage following coagulation or filtration.

In the storage of water in reservoirs or in large settling basins, aération is frequently of benefit in reducing stagnant conditions and microscopic growths by delivering a water rich in oxygen and low in carbon dioxide.

At one time it was believed that aération improved the hygienic quality of water. This is not so. Analyses have shown that bacteria

are not destroyed to any extent. Neither are appreciable changes wrought in the condition of the organic matter in the water. The time elapsing during aëration is too short for this. Oxygen, however, being added to water, the oxidizing capacity of the water is increased and putrefaction, if existent, will cease.

The benefits incident to aëration other than odor or taste removal are in many cases fully as important or even more so. Aëration, therefore, frequently becomes an economic process of water purification even when the reduction of tastes and odors is only of secondary significance.

Principles of Aëration. — The solution and precipitation of gases by aëration are governed by the gas laws, the principles of which are well established. The agencies effecting the removal of non-gaseous odors and tastes, however, are as yet incompletely understood. They are probably in many ways analogous to those causing the precipitation of gases. Familiarity with the gas laws is, therefore, of great assistance in the interpretation of aëration phenomena. Particular attention should be paid to Dalton's expansion of Henry's law which explains the interchange of gases as taking place in accordance with the partial pressures of the gases in the liquid and gaseous phases. (See Chapter VIII.)

The efficiency of aëration varies greatly with the design of the aëerator. Theoretical considerations and practical experience permit formulation of the following general statements that should be appreciated in aëerator design.

- (1) *The ^{greater}
smaller* the concentration of the gas in the water and the *smaller* its volume in the air the more rapid is its *precipitation.*
greater solution.
- (2) *The greater* the surface of water exposed to the air in a unit of time the more effective are the absorption and sweeping-out processes.
- (3) *The greater* within limits the change of air in contact with the water the faster does aëration proceed.
- (4) *The longer* the time of aëration the more far-reaching are the results.

When boiled down, these rules are in fact merely a restatement of Dalton's law from several viewpoints. Their general applicability to processes of aëration is supported by experiments some of which are summarized in the following paragraphs.

Rate of Aëration. — In 1907 some experiments were made by the author, assisted by Mr. Melville C. Whipple, at the Polytechnic Institute of Brooklyn, N. Y., for Messrs. Hazen and Fuller in connection with their report to the New York Board of Water Supply. Deaërated

water was exposed to the air in various receptacles and caused to fall through the air as drops. The rate of oxygen absorption during varying intervals of aëration was then determined. Water containing carbonic acid was similarly tested, and the rate of decarbonation was ascertained. Water charged with sulphureted hydrogen, oil of peppermint, and other essential oils was also studied.

In brief, it was found that an exposure of water to the air in drops for a period of one second increased the dissolved oxygen from 0 up to about 75 per cent of saturation, and an exposure of two seconds raised it further to about 90 per cent.

Carbonic acid was reduced after exposure in drops, as shown by the following figures, which give the quantity left in solution after different intervals of time.

TABLE 97
CARBON DIOXIDE LEFT IN SOLUTION AFTER AËRATION

	Carbon Dioxide (Parts per Million)			
At the start.....	5.0	10.0	25.0	50.0
After 0.5 second.....	4.1	6.9	13.8	23.4
" 1 "	3.5	5.3	9.3	14.0
" 2 "	3.0	4.1	6.2	8.5
" 5 "	2.5	3.0	3.8	4.5
" 15 "	2.1	2.1	2.1	2.1

It will be observed that the greater the concentration of the gas in the liquid phase the greater was the rate of decarbonation.

Sulphureted hydrogen was reduced as follows:

TABLE 98
REDUCTION IN SULPHURETED HYDROGEN AFTER AËRATION

Time	Sulphureted Hydrogen (Parts per Million)	Odor
At start.....	15.2	Faint
After 1 second.....	10.2	Very faint
After 1.5 seconds	5.0	Very faint
After 2.0 seconds.....	2.6	None

The oil of peppermint gave a distinct odor when diluted in water to the extent of one part in one million; it could be detected when diluted to one in fifty million. On exposure to the air in drops the odors decreased as follows:

TABLE 99
REDUCTION IN ODOR AFTER AËRATION

	Odor of Peppermint		
	1	2	3
At start.....	Distinct	Faint	Very faint
After 1 second....	Distinct	Faint	Very faint
After 1.5 seconds...	Distinct	Very faint	None
After 2.0 seconds....	Faint	None	None

Similar series of experiments have been made for a number of years by students in Sanitary Engineering in the Harvard Engineering School. From the results of these experiments the following figures are taken to show the influence of the size of drop upon the rate of decarbonation and by analogy on aëration as a whole. The reduction in carbon dioxide, together with the changes in hydrogen ion concentration, are recorded in Table 100.

TABLE 100
EFFECT OF SIZE OF DROP ON AËRATION

	Carbon Dioxide (p.p.m.)		Hydrogen Ion Concentration (pH)	
	6 mm. Nozzle	1 mm. Nozzle	6 mm. Nozzle	1 mm. Nozzle
At the start.....	71	71	5.6	5.6
After 0.25 seconds.....	68	38	5.7	5.85
“ 0.5 “	44	16	5.8	6.2
“ 0.75 “	28	5	5.9	6.7
“ 1.0 “	24	2	6.0	6.9
“ 2.0 “	7	2	6.4	7.0

NOTE: 0.33 second elapsed before the jet issuing from the nozzle broke into drops.

Natural Aération. — As shown in the Chapters on Limnology and Rheology, aération is ever active in nature both in standing and running water. In lakes, ponds, and reservoirs, wind and thermal currents are constantly bringing new surfaces of water in contact with the air but aération is limited to the zone of circulation as diffusion is extremely slow. In brooks and streams, eddies and currents result in repeated exposure of new volumes of water to the air. The more turbulent the stream the greater the aération of its waters. The natural aération that occurs when water flows down the rocky bed of a brook or over waterfalls has been found repeatedly to be of benefit in reducing odors. Mechanical agitation tends to disintegrate the organisms. Odors are liberated and air is absorbed and provides oxygen for oxidation processes. Whereas a similar disintegration of organisms may take place in the pipes of a distribution system, there is no chance for the odoriferous substances to escape; as a result the disintegration of the organisms may intensify the odor of the water.

Artificial Aération. — In the artificial aération of water, exposure of new surfaces must be accelerated in order to secure adequate results in a short interval of time. We may distinguish between three types of aérators.

- (1) *Injection aerators* in which air is blown into the water.
- (2) *Gravity aérators* in which the water drops through perforated pans, flows in a thin sheet over suitably baffled inclined planes, or percolates through porous materials.
- (3) *Fountain aérators* which spray the water into the air through up-turned pipes or nozzles.

Injection Aération. — One of the early instances of artificial aération was the use of injection aérators in the reservoirs of the Hackensack Water Company in New Jersey. Here air was blown into the stagnant layers of the reservoirs through a system of perforated pipes placed near the bottom. Beneficial results were produced. In small reservoirs bad stagnation conditions can sometimes be averted by this means which liberates odoriferous gases, adds oxygen to the water, establishes aërobic conditions and precipitates iron or prevents it from being taken into solution.

Aération by means of diffuser plates, similar to those employed in the activated sludge process of sewage treatment, has been described by Delaporte. At Essex, Ontario, a plate one foot square was placed at the bottom of a ground water reservoir holding about a day's supply. Air was applied at a rate of 5 to 7 cubic feet per minute (approximately 5 cubic feet per 100 gals.) and the results were as follows: the dissolved oxygen increased from 2.0 to 4.5 cc. per liter; carbon dioxide

decreased from 6.6 to 3.9 p.p.m.; hydrogen sulphide present in the wells to the extent of 9.0 to 10.5 p.p.m. was completely removed; as a result the chlorine required to sterilize the water fell from as much as 40 p.p.m. to about 3 p.p.m. At Richmond Hill, Ontario, a similar installation in the sedimentation chamber of a filter plant greatly improved the quality of a surface water supply drawn from a stagnant reservoir. Air-lift pumps offer another example of aëration by injection. Used in connection with iron- or manganese-bearing waters, the air produces marked oxidation and precipitation of these metals. At Memphis, Tenn., air-lift reduces the carbonic acid from 120 to 32 p.p.m.

In a few installations air is injected into water by utilizing the velocity of the water in pipes and conduits to suck in air by an inspirator effect similar to that of a water jet vacuum pump.

On the whole, blowing air into water is a less efficient and more expensive means of aëration than letting the water fall through the air. The method has its place as an emergency measure or where head for gravity or fountain aëration is not available. In cold climates it can also be used to advantage. In sewage treatment by the activated sludge method, injection aëration has developed into a process of great importance.

Gravity Aërotors. — When a small head is available for aëration it is utilized most conveniently by some form of gravity aërotor. The efficiency secured depends upon the relative surface of water exposed to the air and the time permitted for an interchange of gases to take place.

At Cambridge, Mass., the effluent from rapid sand filters passes from flumes over inclined planes studded with "riffle" plates arranged in herring bone fashion which cause turbulent flow. (See Fig. 108.) The planes or aprons are sloped 3 feet on 8 feet and cover an area of 923 sq. ft. The capacity of the aërotor is 14 million gallons daily and the loading is therefore about 10 gallons per square foot per minute, or 3.5 g.p.m. per sq. ft. per ft. loss of head. From 20 to 30 per cent of CO_2 is released and the aromatic odor characteristic of Cambridge water escapes. Three feet of head are lost. At Portsmouth, Va., a cascade of four steps 13 inches high is used instead of the inclined plane. About 40 per cent of CO_2 is liberated. The spillway of the Croton Dam shown in Fig. 109 is an impressive example of a cascade aërotor.

At South Norwalk, Conn., an aërotor precedes slow sand filters largely for the purpose of replenishing the oxygen supply of the stagnant surface water used. The aërotor is a steel box 9.25 feet long, 6.5 feet wide, and 4.0 feet high. It is housed in a concrete tower. The bottom of

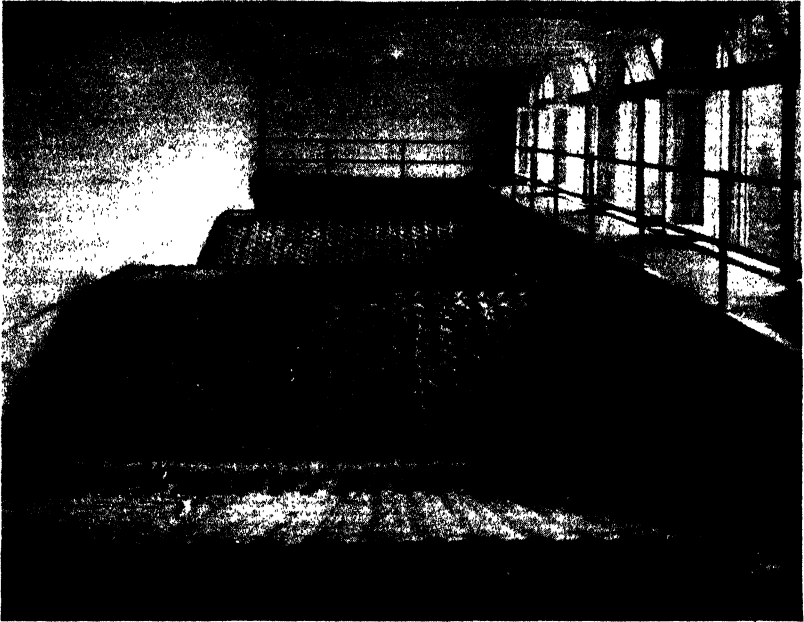


FIG. 108. — Gravity Aëerator at Cambridge, Mass.

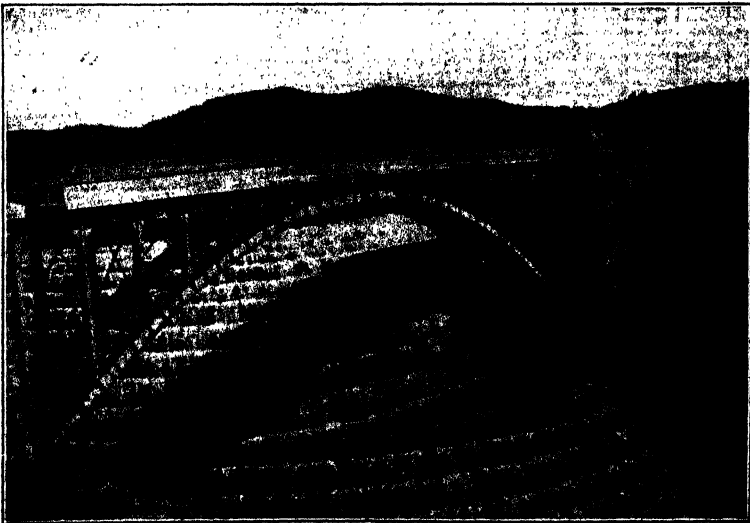


FIG. 109. — Spillway, of Croton Dam, showing Natural Aëration..

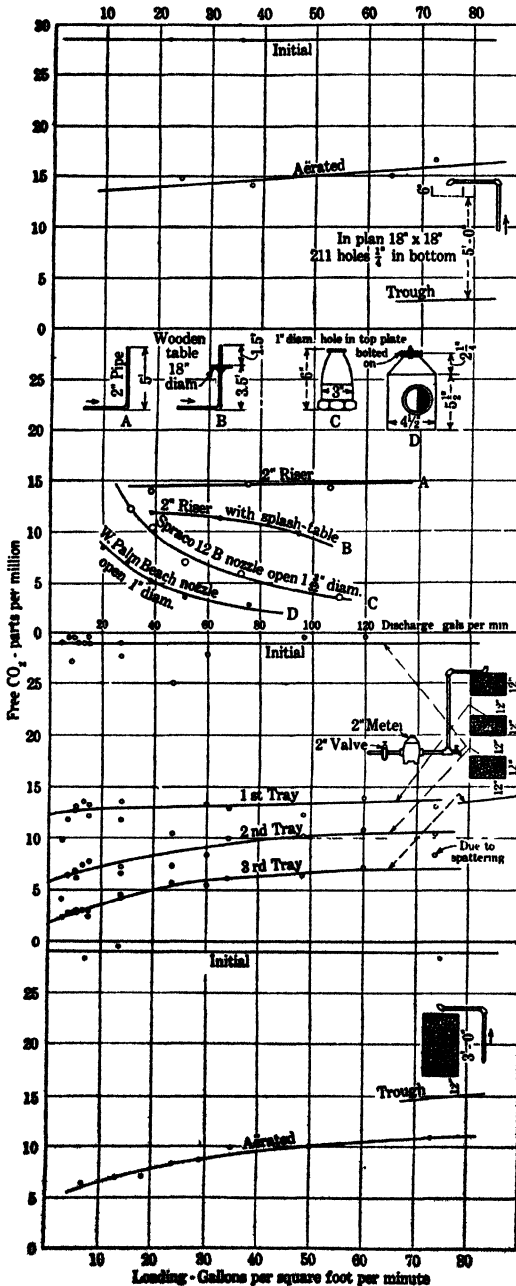


FIG. 110. — Donaldson's Experiments on Aëration. After *Engineering News-Record*. Vol. 90, 1923, p. 874.

the box, tray, or pan is perforated by 6836 holes $\frac{3}{16}$ inch in diameter and spaced 1 inch on centers. The water passes downward through these holes and breaks into drops 6 inches below the orifices. Experiments made with this type of aëerator at Lawrence, Mass., showed that water devoid of oxygen could be saturated in a fall of $3\frac{1}{2}$ feet. The aëerator treats 2.5 million gallons daily, thus carrying a load of 29 gallons per square ft. per minute or about 4 g.p.m. per sq. ft. per ft. loss of head. The effect of housing aëra-tors of this type is illustrated by results obtained from two per-forated-pan aëra-tors at Portsmouth, Va. One of the pans is situated over an open tank, the other in a covered basin. The open aë-rator reduced the CO₂ by 40 per cent, the covered one by 30 per cent. The holes in the pan are $\frac{1}{4}$ inch in diam-eter and the height of fall is 8 feet.

In the removal of iron from public water

supplies, aération can be accomplished together with contact treatment of the water by allowing it to trickle slowly downward through porous materials. The tricklers may be constructed of broken stone, coke, shavings, brick, laths, or other substances. It is important that they be well ventilated. Examples of different types of tricklers are especially numerous in German water supplies. The results that can be obtained by this method of aération as well as by other gravity aérateurs are illustrated by the experiments of Donaldson at Memphis, Tenn., which are shown in Fig. 110. In comparing the efficiencies of different aérateurs the loadings and head losses must be taken into account. Although Donaldson's results show merely the removal of carbon dioxide, it stands to reason that the liberation of odors and tastes and the absorption of oxygen take place at proportionate rates.

Fountain Aérateurs. — Where greater heads are available, aération is secured most effectively by the use of suitably designed fountain aérateurs in which the water is projected into the air, preferably in fine drops, and then falls into a collecting basin.

The development of fountain aérateurs is illustrated in Figs. 111 to 114. Figure 111 shows a simple upturned pipe used at the old Ludlow filters of the City of Springfield, Mass. The head available was small and the water flowed as a sheet rather than a spray. Figure 112 shows the fountain aérateur of the Little River filters of the same city. The head used in aération is much greater and the water is sprayed into the air through multiple openings in a dome casting that caps the fountain. The use of nozzles is shown in Figs. 113 and 114, photographs of the Ashokan and Rye aérateurs. Here the jets issuing from the nozzles break up into fine sprays consisting of small drops which under high heads are almost atomized. The Ashokan aérateur has a nominal capacity of 400 million gallons daily and discharges water through 1600 1½-inch nozzles. The aérateur covers a floor space of 77,000 sq. ft. Its loading is 3.6 g.p.m. per sq. ft. or .15 g.p.m. per sq. ft. per ft. loss of head for an operating head of 24 feet.

The efficiency of several types of fountain aérateurs in removing carbon dioxide has been shown in Fig. 110. The head utilized is commonly the controlling factor in well-designed aérateurs of this type. Fountain aération is used below many dams to aérate the water discharging into aqueducts leading to the city or emptying into other reservoirs. The aérateurs of the New York Water Supply, two of which are shown in Figs. 113 and 114, are especially noteworthy because of their size. Fountain aérateurs are also employed in connection with filter plants, both before and after filtration, as at Providence, R. I., and Albany, N. Y. At Fort Worth, Texas, aération precedes coagulation and fil-



FIG. 111. — Aëerator at the Ludlow Filter, Springfield, Mass.

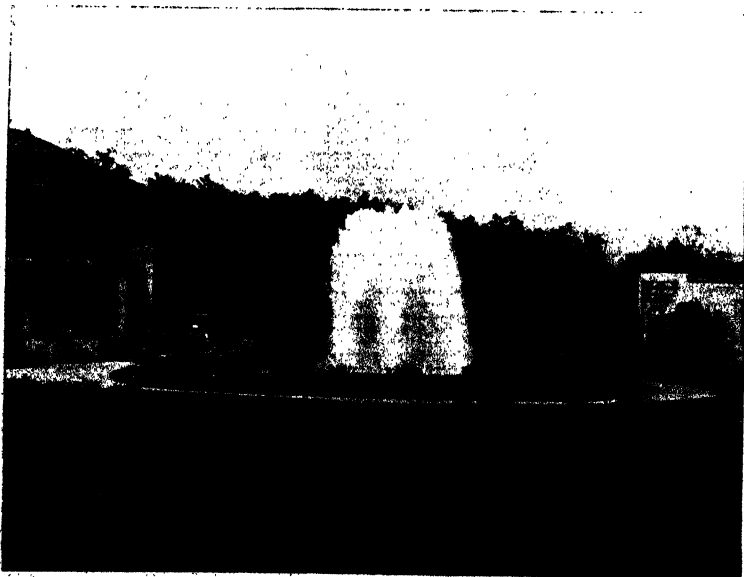


FIG. 112. — Aëerator at the Little River Filter, Springfield, Mass.



FIG. 113 — Ashokan Aerator Catskill Water Supply of New York
Courtesy of Allen Hazen.



FIG. 114. — Aerator at Rye Pond. Borough of the Bronx, New York City.

tration. The aëerator is situated over the coagulation basin and consists of 64 "Spraco" nozzles operating under a 17-foot head and tapped into five cast-iron feeders. Mahlie reports a series of analyses of the water before and after aëration. Four of them are shown in Table 101. They throw some light on the results that may be expected by aëration of stored surface water.

TABLE 101
RESULTS OF AËRATION AT FORT WORTH, TEXAS
After Mahlie
Unless otherwise stated Results are Parts per Million

	Sept. 15, 1922		Jan. 3, 1923		Oct. 8, 1923		Nov. 1, 1923	
	Raw	Aërated	Raw	Aërated	Raw	Aërated	Raw	Aërated
Temperature °F.....	77	77	51	51	76	74	59	57
Turbidity.....	26	25	20	20	35	35	130	130
Color.....	5	5	3	3	5	5	10	10
Odor.....	3e	2e	1e	1e	3e	2e	2e	2e
Dissolved Oxygen.....	4.8	7.8	10.7	11.2	6.3	8.2	8.2	9.6
Per Cent Saturation.....	57	94	96	100	75	95	81	93
Free Ammonia.....	.012	.014	.004	.006	.026	.026	.016	.016
Albuminoid Ammonia.....	.144	.136	.128	.128	.128	.132	.144	.146
Nitrites.....	.001	.001	T	T	.001	.001	.001	.001
Nitrates.....	.12	.12	.12	.1213	.13
Oxygen Consumed.....	3.5	3.4	3.9	3.9	3.1	3.3	4.3	4.3
Remarks.....	Water has taste		Water has no taste		Water in transition; very slight taste at first, finally no taste.			

Deferrization plants are often equipped with fountain aërotors which may be arranged to distribute water over coke or other contact beds. Too much aëration is sometimes a detriment to deferrization processes.

In sewage disposal, fountain aëration is a valuable adjunct to sprinkling or trickling filters.

Aërating fountains are capable of artistic treatment and add to the attractiveness of reservoirs and water purification works. The enjoyment attendant on watching falling water seems to be instinctive.

Aërating Nozzles. — A number of different designs have been evolved as a result of the increased use of nozzles for aëration. Some typical nozzles are illustrated in Fig. 115. The New York nozzle is rifled to give the water a whirling motion as it issues into the air. This gives the cone of water a wider angle, breaks the water into small drops, and

causes an intimate mixture of air and water. Similar results are obtained in the West Palm Beach nozzle by inducing a rotary motion as the water enters the bottle-shaped nozzle. In the Berlin nozzle two jets impinge upon one another and produce a finely divided spray.

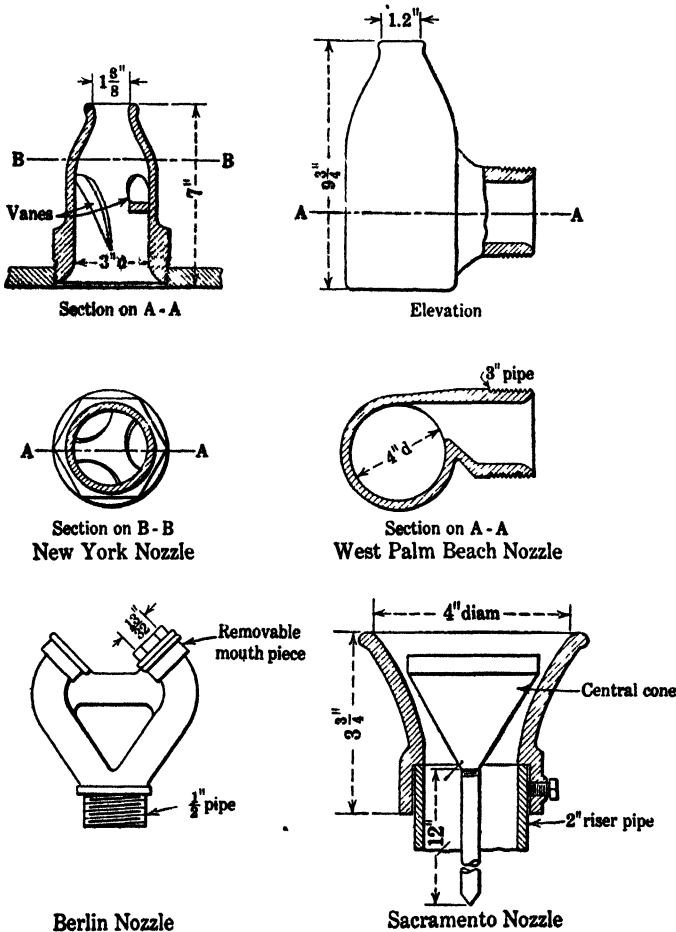


FIG. 115. — Aërating Nozzles.

Berlin Nozzle, *Engineering News-Record*, 1923, Vol. 91, p. 427. Sacramento Nozzle, *Engineering News-Record*, 1922, Vol. 89, p. 386. New York Nozzle and West Palm Beach Nozzle. *Courtesy of Allen Hazen.*

The Sacramento nozzle was designed for low heads (4–5 feet) and contains a central cone which rides on the jet and automatically changes the nozzle opening as the head rises or falls.

The New York and West Palm Beach nozzles were tested by Donald-

son who obtained the results shown in Fig. 110. Mechanical agitation incident to nozzle aëration will destroy the more fragile plankton organisms and will remove the odors due to the essential oils thus liberated.

FILTRATION

The study of water filtration from the standpoint of the microscopist involves a consideration of both the effect of filtration upon microscopic organisms and the influence of algæ upon filtration. With modern methods of reservoir control the latter is often the more important of the two.

When water contains few algæ it can be filtered by either slow sand filtration or rapid sand filtration. Usually the choice of the method is determined by other considerations than the presence of algæ except when the number of organisms is great.

Compared with bacteria and particles of clay, most algæ are relatively large. As a result they are strained out of the water in close proximity to the surface of the sand and become an integral part of the slimy layer called "Schnutzdecke" by German observers who first studied its composition and activity. The accumulation of microscopic organisms in this layer or mat tends to clog the filters and results in a rapid loss of head.

The odor- and taste-producing substances of algæ that are set free during growth or more particularly following destruction of the cells are in part absorbed by the sand grains as evidenced by the marked odor of sand that has filtered water rich in microscopic life; in part they pass through the filter and will cause complaints unless removed by aëration.

Growth of Algæ on Open Filters. — The first slow sand filters built in Europe and America were beds exposed to light and air. As such they were subject to the same algæ growths as shallow open reservoirs, with the exception that the sand surface corresponding to the reservoir bottom was cleaned more frequently. In higher northern latitudes the advantage of covering the beds to prevent ice troubles during the winter months was soon manifested, and with it came a reduction in algal troubles. In milder climates covered beds have been much used for sanitary reasons and for the concomitant prevention of plankton growths. Rapid sand filters have not given much trouble from the growth of organisms because of the frequency with which the beds are washed and, to a less extent, because the relatively low cost of housing them has usually permitted their being covered.

When water is filtered through open sand filters that are operated

continuously, algæ grow upon the sand surface. That this is a growth and not a mere accumulation of strained-out organisms was shown by some experiments made by the author many years ago at Chestnut Hill reservoir.

An experimental filter became so clogged after running for 25 days that it was necessary to scrape the surface of the sand. Microscopical examinations showed that over each square centimeter there were 2,500,000 *Tabellaria* and 1,000,000 *Synedra*, besides many other microscopic organisms. Calculations from the analyses of the raw water indicated that during the 25 days when the filter had been in operation only 150,000 *Tabellaria* and 20,000 *Synedra* were removed from the water by each square centimeter of the filter. The difference between the two sets of figures, therefore, represents the growth of organisms upon the sand. Samples of scum taken from various filters in practical operation have yielded microscopic organisms in numbers that range from a few thousand to several million per square centimeter of surface area. The presence of these organisms aids filtration in a certain sense by forming over the sand a tenacious surface scum that constitutes an excellent filtering medium. This *Schmutzdecke*, however, is built up to a sufficient degree even in the absence of algæ and accumulations of organisms upon the sand are, on the whole, likely to do more harm than good. They cause the filter to clog more quickly than it otherwise would, and, therefore, increase the cost of operation. In the presence of sunlight algæ sometimes interfere with filtration in another way. In the course of photosynthesis and respiration they produce oxygen and CO_2 . When their growth is vigorous the amount of gas liberated by them sometimes becomes so great that masses of the organisms are lifted from the sand layer and float to the surface of the water. Spots of sand are then left bare and the water filters through them more rapidly than elsewhere in the filter and at a higher rate than desirable for good filtration. Filtration then becomes imperfect. It seems probable, also, that decomposition of the organisms at the surface affects the filtered water unfavorably. When filters are covered, chlorophyllaceous organisms do not grow on the sand surface. Those which are found there represent accumulations of cells strained from the raw water.

Experiences at Hamburg and Antwerp. — Dr. Adolph Kemna has made systematic studies of the algæ found in the *Schmutzdecke* whenever a filter bed at Hamburg was scraped. A summary of his work may be found in a discussion by the author in the *Transactions of the Am. Soc. C. E.*, Vol. XLIII, p. 318, from which the following is quoted, substantially as there recorded.

The organisms that develop over the surface of a sand filter may be divided, for practical purposes, into three classes: those which form a matting upon the sand; those which are attached to the sand but extend upward in filaments or sheets; and those which are free-floating in the water. Perhaps it would be better to say that the organisms are found in these three conditions of existence because the same organism is sometimes found now on the sand and now above it.

The effects of these three classes of organisms upon the operation of the filter are not the same. The most important effect is that produced by those organisms which form a matting upon the sand. The diatoms and the unicellular algæ are here chiefly concerned. By their growth they form a more or less gelatinous film upon the surface, and as this film becomes denser, the rate of filtration is retarded until finally it becomes necessary to scrape the filter. The algæ that grow erect upon the sand do not thus clog the filter. On the contrary, they prevent clogging to some extent. Their waving, interlaced threads, acting in the nature of a preliminary strainer, remove from the applied water some of the suspended matter that would otherwise collect on the sand. This action continues as long as the plants are in good condition and as long as the evolution of gas is sufficient to cause flotation. When they begin to decay or become overloaded with foreign matter they settle to the bottom and help to clog the filter. Kemna found that at Antwerp *Hydrodictyon* was the most effective organism in this process of preliminary straining. The free-floating forms have little influence on the rate of filtration as long as they remain in suspension, although, to some extent, they too play a part in the preliminary clarifying process. But ultimately most of them reach the surface of the sand and help to clog the filter.

During the course of the year the character of the flora changes. This change is often gradual, but at times it becomes very rapid. Kemna has noticed that during the time when certain organisms are rapidly disappearing from the sand the efficiency of filtration is impaired. He attributes this to the changed condition of the surface film caused by the decomposition of the organisms, but suggests that changes in the bacterial flora may also play an important part. In one of his publications he cites the following interesting experience with *Anabæna*:

During the hot weather of July, 1899, *Anabæna* became abundant over some of the Antwerp filter beds. Knowing the character of this organism and its tendency to impart an odor to the water, he kept a careful watch of the filters, collecting samples of the filtered water twice a day and testing them for their odor and the amount of ammonia they contained. As long as the *Anabæna* remained alive in the water over the sand, the filtered water was satisfactory, but when the organisms disappeared, on the advent of cold weather, the filtered water acquired a bad taste and the amount of ammonia increased.

The studies made at Hamburg and at Antwerp show, with apparent conclusiveness, that when the vegetation over a sand filter is alive, it is a positive aid to the efficiency of filtration, though it increases the cost of operation. Most of the microscopic organisms have a coating which is somewhat gelatinous, and in many cases the gelatinous material is very abundant. The diatoms and other organisms that grow directly on the sand aid in the formation of the surface film on which the efficiency of filtration largely, but not solely, depends. This fact has been known for many years. The surface film develops through bacterial agency on covered filters as well as on open filters, but on the latter its formation is assisted by microscopic organisms.

Examination of Filter Scum.—As an example of the number of organisms that may be found upon the surface of an open sand filter,

Table 102 is taken from the records of an experimental filter at Boston, Mass. The sample was collected in March after the filter had been in operation two months.

TABLE 102
MICROORGANISMS IN FILTER SCUM

	Std. Units of Organisms over 1 sq. cm. of Sand		Std. Units of Organisms over 1 sq. cm. of Sand
<i>Diatomaceæ:</i>		<i>Cyanophyceæ:</i>	
Asterionella.....	278,000	Cyrococcus.....	5,300
Cymbella.....	130,000	Oscillatoria.....	84,000
Diatoma.....	150,000		
Melosira.....	10,000	<i>Protozoa:</i>	
Meridion.....	25,000	Trachelomonas.....	16,000
Navicula.....	7,700	Ciliata.....	5,000
Stephanodiscus.....	6,500	Peridinium.....	4,000
Synedra.....	1,100,000	Tintinnus.....	14,000
Tabellaria.....	2,390,000	Mallomonas.....	800
		Synura.....	6,000
<i>Chlorophyceæ:</i>		Codonella.....	400
Closterium.....	1,200	<i>Rotifera:</i>	
Scenedesmus.....	800	Anuræa.....	800
Protococcus.....	60,500	Polyarthra.....	1,000
Tribonema.....	12,000	Synchaeta.....	8,000
Spirogyra.....	5,500		
Total organisms.....		4,324,500	
Amorphous matter.....		2,300,000	

Growth of Organisms in Covered Filters. — Since the chlorophyllaceous algæ will not develop in the absence of light the organisms of this group found in covered filters represent merely an accumulation of the cells carried into the sand by the raw water. A covered filter may be likened to a covered reservoir and is limited to the same algal troubles. In the dark the green plankton organisms die but nevertheless form, in conjunction with the suspended matter strained out of the water, a surface film upon the sand. In this film bacteria and fungi thrive and produce zoöglöeal masses that, as the name implies, are gelatinous and sticky in nature. When filters were first covered it was believed that the Schmutzdecke would not be sufficiently heavy or active to produce as good results as were obtained in open filters. It was soon discovered, however, that the film was an adequate straining medium.

The absence of a Schmutzdecke during the initial period of a filter run explains in part why filtration is less effective at this time. Besides the passive rôle of straining out suspended matter the living organisms found in the slimy coatings on the sand particles also play an active part in filtration. Given sufficient time they produce in the organic matter carried by the water changes that are in the nature of oxidation processes. They also act upon the bacteria present in the raw water as evidenced by the great reduction in bacterial efficiency when filters are sterilized intentionally by disinfecting agents or, as sometimes happens unintentionally, by bactericidal trade wastes such as acid mine liquors.

In rapid sand filters the time of passage through the sand bed is so short that biological activity is greatly reduced. Filtration, too, would be less effective were it not for the coagulant floc commonly used in rapid sand filter plants, part of which deposits on the sand surface and thus creates an artificial Schmutzdecke.

The most important growths of microscopic organisms in covered filters are caused by those species that develop without the stimulus of sunlight, notably the fungi. Among these are certain members of the family Chlamydobacteriaceæ, or higher bacteria, such as *Crenothrix*, *Leptothrix*, *Sphærotilus dichotomus*, and forms more nearly resembling the molds that are difficult of identification. In the presence of sulphur compounds sulphur bacteria are also capable of growing.

At Cambridge, Mass., *Leptothrix* has been observed almost constantly in the alum solution system and at times has been found growing on the wooden baffles of the sedimentation basin and even on the sand of the rapid filters. Here the growth reduced the length of filter runs. In the alum solution lines *Leptothrix* has occurred in such masses that it has actually interfered with the discharge of the coagulant. The method of control has been regular dosage of the alum lines with strong solutions of hypochlorite of lime supplemented by steaming and flushing.

At Auburn, New York, growths of hydrozoa and slime-producing organisms have been observed on the columns and walls of the slow sand filters and on the beds themselves. Microscopic examination of the slime showed the presence of *Hydra*, *Beggiatoa* and indefinite thread forms together with bacteria. The growth of *Hydra* at times was colored a brilliant pink and occurred principally in cold weather. The presence of *Hydra* in the filter indicates the occurrence in the water of large growths of crustacea, its principal food.

When water containing a great deal of organic matter or many plankton is filtered through slow sand filters oxidation of the decom-

posing matter may exhaust the supply of dissolved oxygen. Iron is then reduced within the filter, goes into solution and appears in the effluent. This has happened at the South Norwalk Filtration Plant and occurred, too, in the Ludlow experimental filters. Under these conditions growths of *Crenothrix* often occur in the underdrains and may even produce clogging by their vigorous development. Aëration then becomes a necessity and must sometimes be used, not only before, but after filtration, in order to keep the oxygen from becoming exhausted. In some instances the quantity of organic matter and algæ has been so great that in spite of aëration the oxygen has become exhausted before the water reached the bottom of the sand bed.

In such cases continuous filtration becomes impracticable and intermittent filtration must be resorted to as in sewage treatment by sand filtration.

Intermittent Sand Filtration.— Perhaps the best illustration of intermittent filtration applied to the purification of water containing algæ was the Ludlow filter at Springfield, Mass., designed in 1905.

The filter was built cheaply for temporary service by leveling a natural mound of sand, so as to obtain a flat area of four acres. This was divided into four beds enclosed by earth embankments. The water was pumped from the reservoir to an aëerator in the center of the plant, from which it was intermittently applied to the different beds. Tile pipes 6 in. and 8 in. in diameter were laid $12\frac{1}{2}$ ft. apart, to serve as underdrains. The sand was 5 ft. deep and had an effective size of 0.30 mm. The aëerator, shown in Fig. 111, was a novel feature of the plant. The filtered water was further aërated by falling from the small drains into a large collecting drain and thence into a wooden flume.

This filter did good service for several years. It was abandoned when the new supply from the Little River became available.

Double Filtration.— Double filtration was tried experimentally at Springfield by the Massachusetts State Board of Health before the Ludlow filter was built. The results of these experiments were summed up in the chemist's report as follows:

Summarizing the discussions upon this point given upon previous pages, it has been found that practically all positive odors were removed by single filtration except during the period of high numbers of *Anabæna* and fermentation of organic matter in the reservoir. During this period single filtration through sand filters at rates of 2,500,000 and 5,000,000 gallons per acre daily failed to remove the odors, but double filtration, even with the secondary filter operating at a rate of 10,000,000 gallons per acre daily, was entirely successful in removing all odors remaining in the water that had passed through the primary filter, although this primary filter was poorly operated at this time. This result was aided by the aëration of the water before passing to the surface of the secondary filter.

Double filtration with liberal aëration has also been used for treating waters of this class, notably at South Norwalk, Conn., and at Mt. Desert, Me.

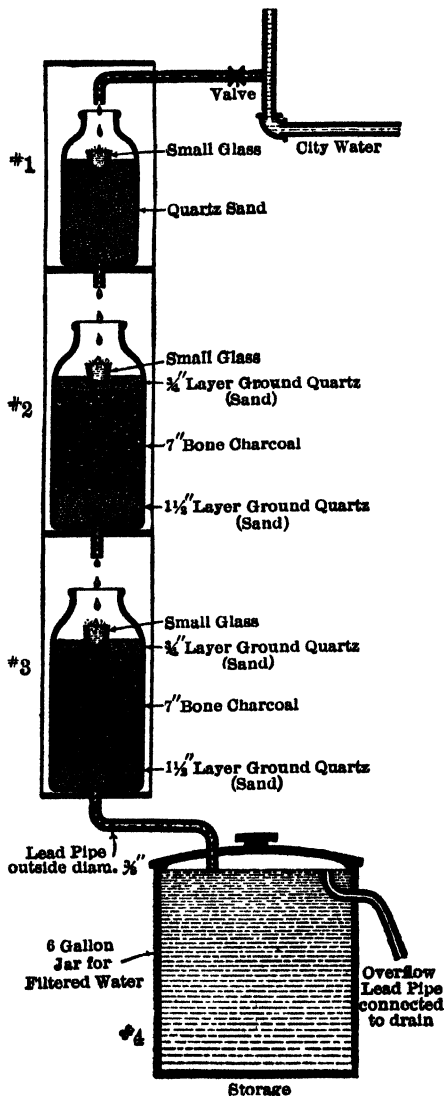


FIG. 116. — Newcomb Household Filter.

A filter of this type, combining aëration and decolorization with charcoal, was much used at Springfield, Mass., in the days of the old Ludlow Supply.

rendered objectionable by the accumulation of bacteria and decaying organic matter. In certain cases the use of sand and charcoal with liberal aëra-

House Filters. — The use of house filters for removing microscopic organisms was at one time quite common. Some of these filters give reasonably satisfactory results if properly cared for, but for sanitary reasons their use is generally not to be recommended. If these filters are not kept scrupulously clean, bacterial slimes will develop and the water may show high bacterial counts and possess disagreeable odors.

Porcelain or stone filters, known as candle filters, such as the Boston filter, the Pasteur filter, and the Berkefeld filter, remove the microscopic organisms completely, but they do not remove all of the odors produced by them. While they may improve the sanitary quality of the water, bacteria are liable to develop in the very pores of the filters and increase rather than decrease the number of bacteria in the effluent. Filters of this type clog rapidly and yield but little water. Cotton disk filters are much used in communities with water rich in algæ. They remove the suspended matter, and if the disks are renewed regularly and frequently they are reasonably clean. Charcoal filters remove the odors as well as the organisms, but are rendered

tion of the water gives reasonably satisfactory results. The Newcomb filter shown in Fig. 116 was used at Springfield when the Ludlow supply was in operation. A filter of this type can be assembled by any ingenious householder, and if he combines cleanliness with ingenuity all will be well. In general, methods of house filtration prove expensive and disappointing.

Resistance to Filtration. — The most important hydraulic effect of algæ upon filtration is the production of increased resistance of the water to passage through the sand. This manifests itself in a rapid building up of the head lost in filtration and results in reduction of the length of filter runs. The cost of cleaning the beds is increased whether scraping and washing are employed as in slow sand filters or backwashing as in rapid sand filters.*

For many years Dr. Houston has studied the resistance to filtration of the many waters under his supervision in the London Water Supply. In order to obtain comparable figures and for the purpose of predicting the behavior of different waters, Sir Alexander has adopted the following method of measuring resistance to filtration.

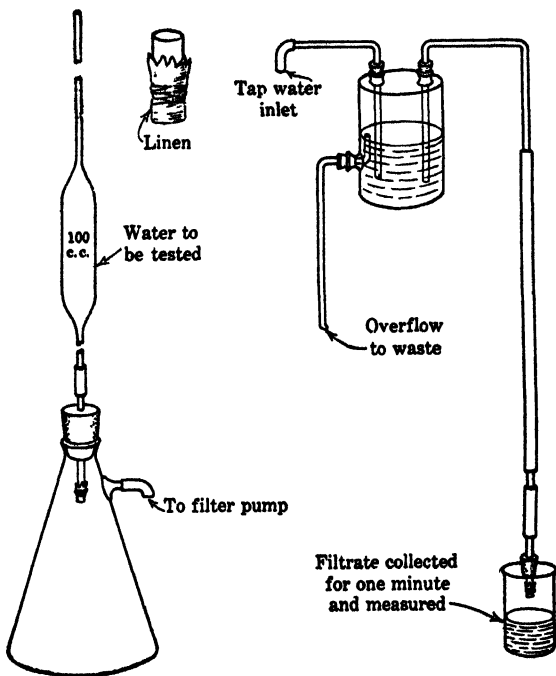


FIG. 117. — Apparatus for the Determination of Resistance to Filtration. *After Houston.*

A known amount of water (100 cc.) is filtered through four layers of superfine linen (96 meshes to the linear inch) that has been moistened and secured by means of a rubber band over the end of a $\frac{1}{4}$ -inch glass tube. Filtration is hastened by suction as shown in Fig. 117. Practically all the suspended matter in the water is retained by the linen.

* At Cambridge, Mass., the runs of mechanical filters were almost doubled by destroying with copper sulphate growths of blue-green algæ in one of the supply reservoirs. The removal of diatoms was of less far-reaching effect.

The tube is next attached to a filtering apparatus in which water can be passed through the linen under a constant head of about 5 feet. This is done and the amount of water passing in one minute is collected and measured. The filtrate collected is an inverse measure of the resistance to filtration. The greater the amount the smaller the resistance.

A convenient set-up for this test in which ordinary tap water is used is included in Fig. 117. The rate of filtration through linen that has passed 100 cc. of clear water varies from 200 to 300 cc. per minute, while the coated linen obtained from water that is badly affected with algal growths may yield no measurable filtrate during this time. Between these two extremes lie most of the other possibilities.

A typical series of results obtained by Houston is illustrated in Fig. 118. This diagram records the seasonal variation in resistance during 1915 to 1916 of the Sunbury Reservoir water compared with raw Thames water and laboratory tap water. It also shows the striking correlation between the laboratory determinations

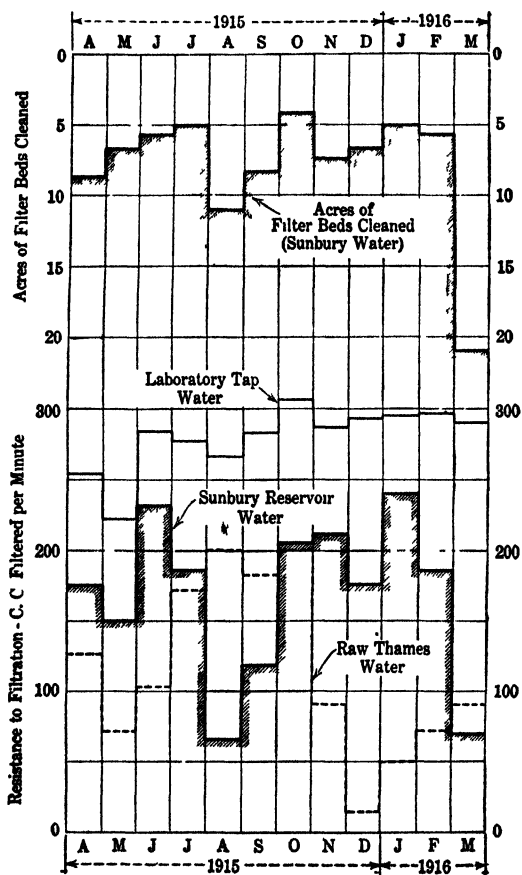


FIG. 118. — Resistance to Filtration of Sunbury Reservoir Water. After Houston.

of resistance to filtration and the number of acres of filter bed cleaned as a result of actual slow sand filtration of this water. The seasonal effect of algal growths is evident. The filtration characteristics of different types of waters are illustrated in Fig. 119 which shows an array of the average resistance to filtration of the various London water supplies and their sources during the decade 1915 to 1925. On the whole the benefits of controlled storage to waters that are to be filtered

are apparent. The low values of the water derived from the West Middlesex reservoirs are explained by the large algal growths to which they are subjected.

In connection with his work on resistance to filtration Dr. Houston has made extensive use of the centrifuge for the concentration of samples and of photomicrographic records for comparative studies (see Chapter V).

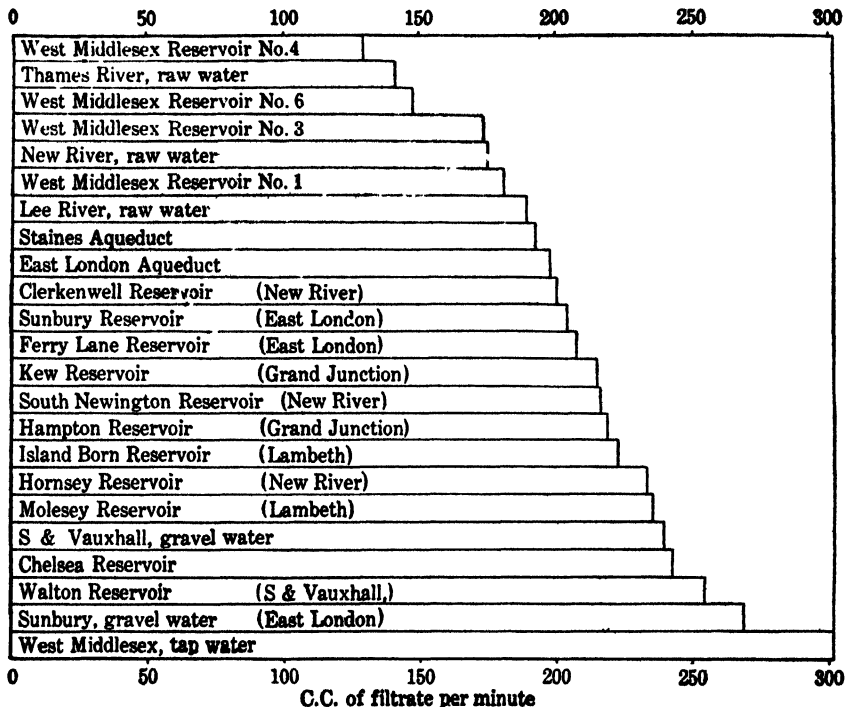


FIG. 119. — Resistance to Filtration of Prefiltration Waters. London Metropolitan Water Board. 10 Year Average, 1915 to 1925. *After Houston.*

Houston's method of determining resistance to filtration provides a laboratory check upon the control of pre-filtration waters. Used in conjunction with microscopic analyses it establishes comparative values of the influence of algæ on filtration and the economics of reservoir control. It indicates necessity for reservoir treatment and helps to explain filter behavior. Where several reservoirs can be drawn upon it is a test that permits the choice of that water which will yield the longest filter runs.

Control of Algæ versus Purification. — As stated in the beginning of this chapter, neither control alone nor purification alone will solve in the most economical fashion the problems of algæ in water supplies.

In planning water works, swamp drainage, reservoir cleaning, arrangement of intake facilities, use of algicides, and employment of purification processes must be considered jointly, as well as separately, in order to develop economically a supply in which the nuisances due to algal growths will be reduced to a minimum. Hazen and Fuller's studies of the Catskill supply of New York City are exemplary of this method of approaching the problem. In operating water works likewise, supervision of catchment area, operation of reservoirs and control of purification methods must be a unified and balanced undertaking to insure success. Laboratory control is essential and freedom from algal troubles will vary in proportion to the chemical and biological skill of the laboratory staff and the coöperation given them by the engineering members of the bureau. The London laboratories under Houston and the New York laboratories under Hale have set a standard of conduct in this respect.

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CHAPTER XV

MICROSCOPIC ORGANISMS IN WATER CONDUITS

Comparatively little has been written in this country upon the biology of aqueducts and pipes. Yet it is an important problem both in its sanitary significance and in its hydraulic aspects. The conditions of existence in the swift-flowing waters of hydraulic conduits are quite different from those obtaining in lakes, ponds and reservoirs or even in streams. The large surface area wetted by the water, the rapid passage of the water through the conduits, the changing velocities, and the exclusion of light in covered aqueducts and in pipes bring about a new alignment of life adapted to the peculiar environment thus created. The time of passage being short, plankton life does not develop to any extent. Fragile organisms indeed are broken up in the turbulent water and the phytoplankton is destroyed in the absence of light. Many organisms settle out in the "dead ends" of distribution systems and quickly go to pieces. Attached organisms predominate, with free-swimming forms seeking shelter in the quiet recesses created by the sessile growths. If the conduits are covered, chlorophyllaceous organisms die off, but attached fungi and animal forms persist and sometimes develop in astonishing bulk. This new alignment of life results in new problems of great interest to the aquatic microscopist.

Reduction of Plankton in Pipes. — The changes that take place in the plankton during passage through water pipes are illustrated by analyses obtained from the Boston distribution system. (Table 103.) Samples were taken from the two distributing reservoirs (Chestnut Hill and Brookline) and from two taps, one at Park Square, five miles from Chestnut Hill, and the other at Mattapan, eleven miles from this reservoir. The results are averages of weekly observations for five years (1891 to 1895).

The greatest reduction did not occur near the reservoirs, where the pipes were large and the currents swift and constant, but at the extremities of the distribution system, where the pipes were smaller and where during the night the velocities were reduced.

The observations showed that during the winter, when there were comparatively few organisms in the water, the reduction in the pipes was much less than during the summer, when organisms were more abundant. During the six months of the year, from November to

TABLE 103
REDUCTION OF PLANKTON IN BOSTON DISTRIBUTION SYSTEM

	Number of Standard Units per cc.	
	Organisms	Amorphous Matter
Chestnut Hill Reservoir.....	248	222
Brookline Reservoir.....	215	212
Tap in Park Square.....	189	190
Tap in Mattapan.....	81	105

April, there was a reduction of 44 per cent in organisms and 24 per cent in amorphous matter in about 6 miles of pipe; while during the six months from May to October the reduction was 62 per cent for the organisms and 53 per cent for the amorphous matter. It is worth noting that the reduction in organisms was greater than the reduction in amorphous matter.

Not only are the microscopic organisms and amorphous matter reduced in the pipes, but the bacteria also tend to change in number, increasing at certain seasons of the year and decreasing at others. This fact has been observed in many cities. In the pipes of the Boston Water Works a decrease occurs throughout the colder months. In the summer, however, when the temperature of the water is high and when the organisms in the water and those growing in the pipes are passing rapidly through stages of growth and decay, there is a considerable increase. This is shown in Fig. 120.

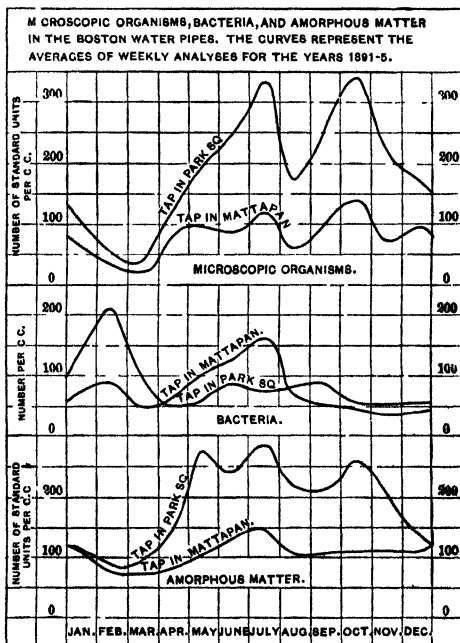


FIG. 120. — Seasonal Variation of Microscopic Organisms During Passage through Water Pipes. Boston, 1891 to 1895.

When water is disinfected the number of bacteria usually increases slightly in the distribution system. Sometimes very large growths of bacteria occur. These are known as after-growths and are believed to be due to rapid multiplication of those organisms that have survived disinfection and find in the water a suitable environment for growth.

The nature of the microscopic organisms showing the greatest reduction in the pipes is evident from the following detailed study of the Boston examinations for the years 1892 and 1893.

TABLE 104

REDUCTION OF PLANKTON GROUPS IN DISTRIBUTION PIPES BETWEEN PARK
SQUARE AND MATAPAN, BOSTON, MASS.

Average for the years 1892 and 1893

Cyanophyceæ.....	54 per cent
Chlorophyceæ.....	57 "
Diatomaceæ.....	58 "
Protozoa.....	64 "
Miscellaneous.....	58 "
Plankton of all kinds.....	56 "

Cause of Reduction of Plankton in Pipes. — Questions naturally arise as to the cause of this reduction of plankton in the pipes. They may be considered under the following topics: sedimentation, disintegration, darkness, and consumption by other organisms.

Most of the plankton organisms are heavier than water. Some always settle in quiet water, and they do so in pipes whenever the velocity of flow is reduced to a certain point. Others, which in ponds usually rise to the surface on account of the gas bubbles which they contain, will settle in the pipes when the pressure of the water, by increasing the solubility of gases, has deprived them of their buoyancy. In dead ends the organisms and particles of amorphous matter often accumulate and form deposits upon the bottom of the pipes. They also tend to deposit on up-grades. Thus it is often found that the water from the high points of a distribution system contains fewer organisms than that from the low points. This has been noticed in high buildings, where the difference between the water on the upper stories and that on the lower floor is often considerable.

Many of the common organisms are very fragile. Even slight agitation of the water will break them up. This is particularly true of protozoa, such as *Synura* and *Uroglenopsis*, but it also happens to the siliceous cells of some diatoms.

Most of the organisms found in surface waters are accustomed to live in the light. Many of them cannot persist in the absence of light.

When they enter the dark pipes they are liable to die and decompose. This is particularly true of some of the chlorophyllaceous organisms that are abundant in the summer. Microscopical examination of samples from the service taps has often revealed decaying plankton organisms swarming with bacteria. The color of the organisms often changes in the pipes of a distribution system. For example the color of *Tabellaria* and other diatoms may be yellowish-brown in the reservoir but greenish-brown in the pipes. Death and absence of sunlight are responsible for these changes.

Another important consideration in the reduction of plankton in pipes is the fact that in many surface water distribution systems the pipes are covered with growths of sponge and other attached organisms that depend for their food material upon the plankton of the water. If the growths are abundant, the removal of plankton from the water by this means may be considerable.

Temperature Changes in Distribution Pipes. — The temperature of water changes during its passage through the pipes of a distribution system. The nature of these changes is shown by Fig. 121, where the curves represent the averages of weekly temperature observations for five years at Chestnut Hill reservoir and at two taps, one at Park Square, 5 miles from the reservoir, and

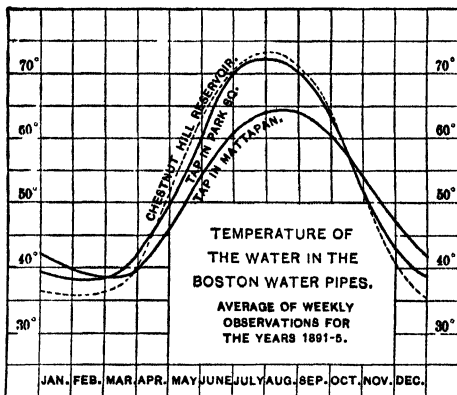


FIG. 121. — Seasonal Temperature Variations of Water in Distribution Pipes. Boston, 1891 to 1895.

the other at Mattapan, 11 miles from the reservoir. During the spring and summer the water grows cooler as it passes through the pipes, and during the autumn and winter it grows warmer. The maximum temperature at Mattapan is never as high as that at Park Square, but the minimum temperature is about the same at both places, though it occurs later in the season at Mattapan.

Temperature changes of this kind can have but little effect upon the plankton, but the smaller variation in temperature is probably favorable to the growth of true pipe-dwelling organisms.

Growth of Organisms in Water Conduits. — Whereas the plankton commonly decreases in water conduits the attached or sessile organisms often find conditions favorable for growth. In open canals and flumes

both plants and animals develop. In covered aqueducts and pipes only light-shunning fungi and animals are able to exist. The flora and fauna of water conduits include a variety of classes of organisms. Covered conduits harbor in particular fungi, sponges, hydrozoa, bryozoa, crustacea, insect larvæ, worms, and mollusks. Open conduits contain more commonly the attached algæ and higher plants. The branching, furry growths caused by the hydrozoa and bryozoa are popularly known as "pipe moss."

Pipe Moss. — In the city of Hamburg the minute animals inhabiting water pipes were studied by Hartwig Petersen in 1876. Ten years later Karl Kræpelin made a more extended study. His observations were of much interest. He found an animal growth, often more than one centimeter thick, covering the entire surface of the pipes. The composition of this growth varied in different places. He gave a list of sixty different species observed. In many places the walls of the pipes were covered with fresh-water sponge, chiefly *Spongilla fluviatilis* and *Spongilla lacustris*. Mollusks were conspicuous, especially the mussel, *Dreissensia polymorpha*. Snails were also numerous. Hundreds of "water-lice" (*Asellus aquaticus*) and "water-crabs" (*Gammarus pulex*) were found at every examination. The material known as "pipe-moss" was common, and consisted largely of the hydrozoön *Cordylophora lacustris* and the bryozoa, *Plumatella* and *Paludicella*.

The tremendous size of some of the bryozoal growths is shown in Fig. 122, a photograph of a growth of *Pectinatella* from the wall of one of the Hartford, Conn., reservoir gate houses. This colony of organisms was twelve inches in diameter. The picture was sent to the author by Mr. Caleb M. Saville, Manager of the Water Works.

The Rotterdam "Water Calamity." — At the time when *Crenothrix* was giving so much trouble at Rotterdam, Hugo de Vries made an extended study of the animals and plants found in the water pipes of that city. His observations were confined chiefly to the pipes and canals which conveyed the unfiltered water of the river Maas to the filter beds. In speaking of one of the canals he said:

The walls were thickly covered with living organisms up to the water-level. They formed an almost continuous coating of varying composition. There were only one or two exceptions to this. In one place, where the water came from the pumps with great velocity, the walls were free from living organisms; and in another place, where there was almost no current, only one living form was seen. There was a section of one of the canals, where a gentle current was flowing, that was a magnificent aquarium. The walls were everywhere covered with white tufts of fresh-water sponge, *Spongilla fluviatilis*. Many of these tufts reached a diameter of 6 or 8 inches, but most of them were somewhat smaller. Between the sponge patches were seated

countless numbers of the mussel, *Dreissensia polymorpha*. Individuals old and young were often seen grouped together in colonies which sometimes extended completely over the sponges. But what most of all attracted attention was a luxuriant growth of the "horn-polyp," *Cordylophora lacustris*. It covered the mussel-shells and occupied all the space between the sponges. The stalks reached a length of an inch or more. On and between the *Cordylophora* swarmed countless numbers of

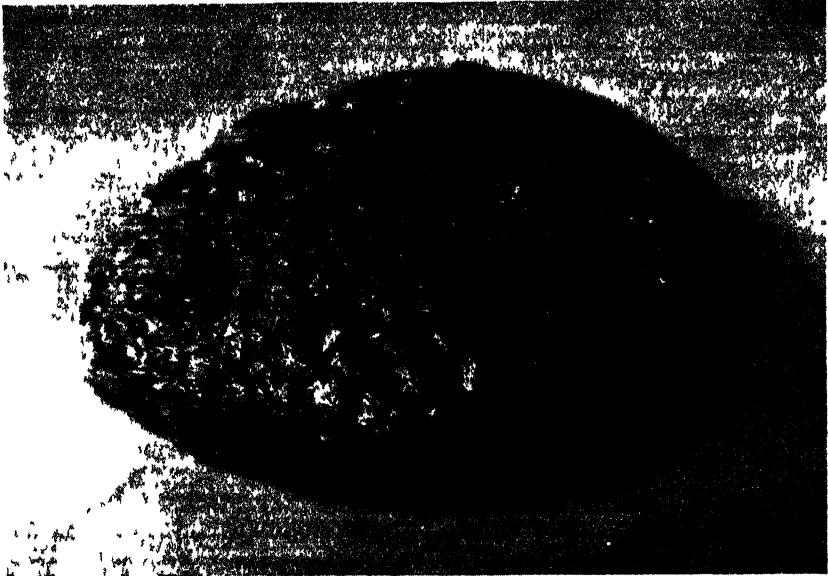


FIG. 122. — Growth of *Pectinatella* from Gate House Wall of a Hartford, Conn., Reservoir. The diameter of this massive growth was 12 inches. *Courtesy of Caleb Mills Saville.*

Vorticella, *Acineteta*, and other protozoa and rotifera. These organisms had no lack of food material, and the absence of light protected them from many foes which, in the light, thin out their ranks. Over all these animals *Crenothrix* was found growing in abundance. The shells of the mussels and the stems of the "horn-polyps" were coated with a thick felt-like layer of these "iron-bacteria." In other localities in the pipes the place of the "horn-polyps" was occupied by the bryozoa, or "moss-animalcules." All of these branching forms were spoken of collectively by the workmen as "pipe-moss."

Boston Experience. — In the summer of 1896, when the pipes of the Metropolitan Water Works were being laid in Beacon Street, Boston, near Chestnut Hill reservoir, a 16-inch main leading from Fisher Hill reservoir to the Brighton district was opened. This afforded an opportunity to examine the material on the inside of a pipe that had been laid ten years. Inspection showed that besides the usual coating of iron rust, tubercles, etc., there were numerous patches of fresh-water

sponge, both *Spongilla* and *Meyenia*, brownish or almost white in color, and about the size of the palm of one's hand. What was most conspicuous, however, was a sort of brown matting that covered much larger areas to a thickness of about $\frac{1}{4}$ inch. It had a very rough surface and, when dried, looked like a piece of coarse burlap. This proved to be an animal form belonging to the bryozoa, known as *Fredericella*. As fragments of it had several times before been observed in the water from the service taps, and as it had been seen growing in some small pipes connected with filtration experiments at Chestnut Hill reservoir, more extended observations were made in different parts of the distribution system.

These brought out the fact that sponges and moss animalcules were well established in the pipes. Many other organisms were also observed. In some places almost pure cultures of *Stentor* and *Zoöthamnium* were found. At other points hosts of different organisms were seen, such as snails, mussels, *Hydra*, *Nais*, and *Anguillula*, *Acineta*, *Vorticella*, *Arcella*, *Amoeba*, countless numbers of ciliated infusoria, and many other forms. The growths were distinctly animal in their nature, but in many places vegetable forms, such as *Achlya*, *Crenothrix*, and *Leptothrix*, were common. The most important class of organisms found, however, was that of the bryozoa, of which *Fredericella* and *Plumatella* were the chief representatives.

Brooklyn Experience. — An interesting experience with pipe moss is on record at the Brooklyn Water Department. In November, 1897, the water in Mt. Prospect reservoir became so filled with *Asterionella* that it was deemed advisable to shut off the reservoir and pump directly into the pipes. This action was followed by the appearance of brown fibrous masses in the tap water. In a number of instances this material stopped up the taps, and even large pipes were choked. The water at the same time had a distinctly moldy and unpleasant odor. The fibrous matting proved to be *Paludicella*. It had been growing on the inner walls of the pipes, and the change of currents and the pulsations of the pump, due to the direct pumping into the pipes, had dislodged it. Systematic and thorough flushing of the pipes materially improved the conditions.

Similar troubles were experienced under the same conditions of operation about 1905. In this case the pipes became so badly clogged by the dislodged growths that it was necessary to call the fire department to aid in flushing out the pipes so that flow might be reestablished.

Food Supply of Pipe Moss. — The fact that many of the organisms that dwell in water pipes depend for their food material upon the algae, protozoa, bacteria, etc., contained in the water has been demonstrated

by the following experiment. Specimens of *Fredericella* and *Plumatella* were placed in a series of jars, some of which were supplied with water rich in microscopic life, while others were supplied with the same water after filtration. All the jars were kept in semi-darkness at the same temperature, and were examined daily. The *Fredericella* and *Plumatella* that had been supplied with filtered water soon began to die, while those in the other jars lived as long as the experiment was continued. Some of the same bryozoa were placed in jars furnished with water from the Newton supply, a ground water almost free from microscopic organisms, and after about a week they died for want of food. Dr. G. H. Parker of Harvard University in a similar experiment on fresh-water sponge obtained the same result. With these facts established, we may confidently affirm that fresh-water sponge, moss animalcules, and similar pipe dwellers will be absent from water pipes where ground water or water that has been effectively filtered is used.

Growth of Crenothrix in Pipes and Wells. — The principal pipe growths so far discussed occur in water derived from surface sources. They are not encountered in pipe lines carrying ground water. Here, however, they are sometimes supplanted by growths that are peculiar to the conditions of existence established by the character of phreatic waters.

The most important denizens of ground water systems are the iron bacteria, and chief among them is *Crenothrix*. As stated before this organism has the power to oxidize certain forms of iron which it precipitates on its sheath or nearby. It thrives best in the dark and is found most frequently in water that contains little or no oxygen but considerable amounts of carbonic acid. The absence of the former and presence of the latter are usually associated with ground water or the stagnant layers of reservoirs and it is here, therefore, that the iron bacteria occur most abundantly. Water rich in carbon dioxide is able to dissolve iron and will carry it in solution as ferrous iron until it is oxidized to the ferric state in which iron compounds become insoluble in water and precipitate. Oxidation can be accomplished either by oxygen dissolved in the water or in its absence by iron bacteria. Iron is thrown out of solution and deposits of iron accumulate. In water pipes these deposits, together with the growth of the organisms themselves, encroach seriously upon the carrying capacity of the conduits.

Crenothrix is known to have occurred in almost all parts of water-supply systems carrying iron-bearing waters. It has been found in wells, pipes, and reservoirs. Tubular wells have become choked with iron deposits resulting from the activity of *Crenothrix*, and distributing

and service pipes have been much reduced in capacity. Except for the reduction in flow, the growth of *Crenothrix* may go unnoticed for a long time, until its disintegration results in an "unloading" of the pipes, and rusty foul-smelling water is discharged over wide sections of the system.

Experiences with Crenothrix Growths. — *Crenothrix* has given annoyance in many water supplies. The "water-calamity" in Berlin first drew attention to its evil effects. In 1878 the water from the Tegel supply became filled with small, yellowish-brown, flocculent masses that settled to the bottom when water was allowed to stand in a jar. The odor of the water and the effects of the iron oxide in washing were decidedly troublesome. *Crenothrix* was not found in Lake Tegel, but was discovered in many wells, in the reservoirs at Charlottenburg and in the unfiltered water of the river Spree.

In 1887 the water supply of Rotterdam, as previously noted, was badly affected with *Crenothrix*. The water was drawn from the river Maas, and after sedimentation was filtered. *Crenothrix* appeared at a time when the system was being enlarged. New filter beds were in use, but the filtered water was conducted through the old conduits, reservoirs, and pumps. In the old conduit, or flume, there were many wooden timbers, and on these *Crenothrix* was found growing in abundance. Upon tracing the source of the trouble it was discovered that the water was imperfectly filtered and that this impure water was the chief cause of the sudden and extensive development of *Crenothrix*.

The way in which water supplies may become infected with growths of iron bacteria is illustrated by the following account of troubles experienced in an industrial plant using ground water from deep wells. When the supply was first put in service the water pumped from the wells was of good quality, and contained only small amounts of iron. A few years later, however, trouble began to be experienced with the clogging of pipes that carried water through the establishment for process purposes. Conditions became worse from year to year until at one time the use of the well water caused the effective area of 6-inch pipes to be reduced to that of 2-inch pipes in the course of a few weeks. An investigation showed that *Crenothrix* was growing luxuriantly in all parts of the system and even covered in a thick mat the outside of water-cooled ammonia condensers. By analysis the well water contained large quantities of carbon dioxide (110 p.p.m.) and iron (6 p.p.m.), but no dissolved oxygen.

A survey of the well area showed that the management of the plant was itself responsible for the change in the quality of the water, which was brought about as follows. The water was gathered from sandy

soil that originally contained but little organic matter, as evidenced by the fact that the ground was almost bare and supported but scant vegetation. For a number of years, however, the plant made a practice of dumping large quantities of organic waste materials upon the well area. These decomposed, impregnated the ground, charged the soil air with carbon dioxide, and used up the oxygen. The carbon dioxide was taken into solution by the percolating water and in time enabled the water to dissolve large amounts of iron. Conditions of existence became favorable to the growth of *Crenothrix* which, once established, developed in large numbers, eventually causing the troubles referred to.

The remedy applied was the removal of the organic wastes and temporary deferrization of the water. It was hoped that in time the ground would purge itself and that the water would then be restored to its original quality.

Growth of Slime-Producing Organisms in Water Conduits. — Slime-producing organisms have been known to grow so abundantly in waste waters as to threaten the operation of industrial processes. The condition has been noted particularly in pulp mills. In the flumes and water boxes of a large sulphite pulp mill, for example, deposits of slime were found that were over a foot in diameter and several inches thick. Examination disclosed the presence of *Beggiatoa*, *Leptomitus*, and *Sphærotilus natans*, the filaments of which formed dense, felted masses containing particles of pulp and fiber. Decomposition proceeded in the center of the masses with occasional loosening of particles which were swept on by the process water and later damaged the product of the mill.

The mill took its process water from a river and discharged its sulphite wastes a short distance below the intake. It was shown that at times the waste water reached the intake, thus increasing the sulphur content of the river water and seeding it at the same time with organisms that had flourished in the mill flumes because of the abundant supply of sulphur. The circle thus established served to perpetuate the trouble in the mill.

Experiments demonstrated the ability of copper sulphate to check the development of the slime-producing organisms when employed in doses as large as 15 pounds per million gallons.

Liquid chlorine in large doses will also destroy these slime-producing organisms, but under conditions such as have been described it would not be as efficacious as copper sulphate, for the reason that considerable amounts of chlorine would be neutralized by organic matter and by sulphites.

Growth of Insect Larvæ in Conduits. — The Hydraulic Power Committee of the National Electric Light Association reports the restriction of flow in some of the power conduits of the Southern California Electric Company by the aquatic larvæ of certain insects such as the caddis worm (*Phryganea*) and the hellgrammite or larva of the salmon fly (*Corydalis cornuta*). These larvæ were found in great numbers growing on the walls of the conduits. The organisms attached themselves to the pipes by means of strong claws at the posterior end of their body and in this position fed on plankton carried past them. The caddis worms were the worst offenders as they increase their bulk by collecting about their bodies a larval case built of sand grains and organic particles fitted together in a silky matrix. The larvæ occurred in dense masses near the upper portals of the tunnels and in the pipes. Growths extended over several miles of conduit. They were most troublesome during spring and early summer, the season of their most rapid development.

Cleaning of the conduits increased their capacity by as much as 8 per cent. The company adopted a practice of cleaning the larger conduits by hand scraping. In smaller pipes, oak brush tied into a rough ball and carried through the pipes by the water was successful in removing much of the growth. It is believed that a smooth coating will reduce troubles due to the growth of aquatic insect larvæ by preventing their gaining a hold on the surface.

Effect of Growths of Organisms in Conduits. — One naturally asks, "What is the effect of these organisms growing in the pipes?" In a certain sense they tend to improve the quality of the water, by reducing the number of floating microscopic organisms; but they themselves must in time die and decay. In all probability, however, very large quantities of decomposing organisms of this type are required to produce noticeable tastes or odors in the water.

Perhaps the greatest objection to pipe growths is the fact that they tend to impede the flow of water. When one considers that a coating $\frac{1}{4}$ inch thick diminishes the cross-sectional area of a 24-inch pipe by 4 per cent and of a 6-inch pipe by 15 per cent, and when one learns that these organisms often form layers even thicker than this, it will be seen that such growths are matters of no little hydraulic importance. Furthermore, fingers of the fresh-water sponge sometimes extend several inches into the water, and the matting of the bryozoa is always rough on account of the stiff branches that are extended by the organisms to secure their food. This roughness of the surface materially increases the friction of the pipe by a considerable but indefinite and constantly changing amount.

Studies of an open concrete canal in the Yakima project of the United States Reclamation Service showed that the seasonal growth of filamentous algæ increased the roughness of the canal expressed in terms of the value n in Kutter's formula from 0.013 to 0.0155. This seasonal increase was found to be quite constant and produced an increased depth of flow of 0.76 feet over that required to deliver the same quantity of water at the beginning of the "growing season."

In January, 1926, gagings of the timber flume of the Hat Creek No. 2 Development of the Pacific Gas & Electric Company which should have a value of n not exceeding 0.013 gave a value of n of 0.0157. In August and September 1922 the value was 0.0248. These high values were due to growths of algæ and moss that lined the sides and bottom of the flume. In the winter examinations it was reported that moss was growing in patches spaced at considerable intervals and covering less than 1 per cent of the wetted area. In the summer studies algæ were present as well as moss. The algæ formed a mat one or two inches thick over the whole wetted perimeter and sent streamers often two feet long into the current.

The carrying capacity of concrete ditches and steel flumes of the San Joaquin Light and Power Company has at times been reduced by 20 per cent, due to algal growths. These growths formed a slimy deposit about $\frac{3}{4}$ inch thick but cut down the capacity more by increasing the frictional resistance to flow than by reducing the area of the channels. Wooden paddles were used to remove the algæ.

Aquatic flowering plants have also caused trouble. Cleaning vegetation from the Miami and Erie canals reduced the values of n from 0.047 to 0.036. Treatment of the banks alone reduced n to 0.042.

The effect of pipe organisms upon corrosion of iron and steel pipe is obscure. It stands to reason, however, that the death and decay of the organisms must at times result in a greater concentration of carbon dioxide in the water which may accelerate corrosion. The adherence of organisms to the pipe wall may furthermore cause a disintegration of the pipe coating either during growth or when the dead organisms are torn from the pipe wall. Uncoated surfaces of the metal then become exposed to the corrosive action of the water. It has been shown on the other hand that deposits on iron pipes frequently prevent corrosion by forming a barrier between the pipe and the water.

Pipe growths, when they become detached from the walls, sometimes cause trouble by clogging service pipes and meters.

Control of Pipe Growths. — Effective control of reservoirs or purification of algæ-laden waters will naturally be attended by reduction or

elimination of pipe-dwelling organisms such as the pipe mosses that are largely dependent upon plankton for food. Deferrization brings about a concomitant destruction of the iron bacteria and it may be stated as a general rule that the purer the water the fewer the troubles with pipe growths.

Temporary fouling of water mains by starvation and subsequent decay of pipe growths has sometimes attended the introduction of plankton control methods or water purification by filtration. Thorough flushing of mains when cleaner water supplants plankton-rich water in distribution systems will often avoid troubles of this nature. At Cambridge, Mass., the change from unfiltered to filtered water was accomplished without untoward experiences. The filters were put into operation during cool weather, and mains were cleaned and flushed about the same time.

Pipe-cleaning tools or scrapers are useful in removing growths. Systematic flushing of pipes at high rates of flow will do much to prevent the accumulation of pipe-dwelling organisms. Dead growths are naturally more easily dislodged than living ones which frequently adhere tenaciously to the conduit walls. Conduit growths including *Crenothrix* can be killed by dosing the water with copper sulphate or with chlorine. Relatively high concentrations of the chemicals are required and it is advisable to isolate the section of conduit to be cleaned, dose it, and flush it thoroughly before it is again put into service. Copper sulphate used in iron pipes where the copper is quickly precipitated when it comes into contact with iron is commonly not as effective as in masonry conduits. Chlorine is better adapted to use in iron pipes. In the presence of reducing agents such as sulphites, however, copper sulphate may be more efficacious than chlorine. *Crenothrix*-infested wells have been successfully treated with chlorine.

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PART II

DETERMINATIVE MICROSCOPY

CHAPTER XVI

CLASSIFICATION OF THE MICROSCOPIC ORGANISMS

The microscopic organisms found in drinking water include the lowest forms of life. Some of them belong to the vegetable kingdom, some to the animal kingdom, while others possess characteristics that pertain to both. There is in reality no sharp dividing-line between the plant and the animal in the low forms of life. Nature's boundaries are always shaded on both sides.

Classification. — Classification of organisms into groups is necessary, but it must be borne in mind that all classifications are artificial and subject to change. The one outlined below is believed to be the most convenient for the work at hand. Several groups, not pertaining to the microscopical examination of drinking water, are omitted.

Nomenclature. — In order to secure greater uniformity in the use of names for organisms, both botanists and zoölogists have adopted codes of nomenclature at international congresses called for the purpose. These codes are based on the principle that the oldest recognizable name of a species should be adopted, and contain rather specific rules and recommendations for the application of this principle. The codes of the botanists and of the zoölogists differ in many minor details and should be consulted in following up problems of nomenclature. The botanists have not yet legislated regarding the nomenclature of the bacteria, but the Society of American Bacteriologists adopted a provisional code at a meeting in Boston in 1919. Since this represents the most consistent practice among bacteriologists up to the present time, it has been followed in this volume.

According to the international rules of nomenclature every individual organism belongs to a species, every species to a genus, every genus to a family, every family to an order, every order to a class and every class to a division. Families are often subdivided into sub-families, tribes, and sub-tribes. All organisms are designated by the name of the genus to which they belong followed by the name (or epithet) of their species.

Both names are Latin or latinized. The generic name is a substantive and begins with a capital letter. The specific name is an adjective and commonly begins with a small letter.

CLASSIFICATION OF THE MICROSCOPIC ORGANISMS

Plants	
ALGÆ Cyanophyceæ (Myxophyceæ) Chlorophyceæ Xanthophyceæ (Heterokontæ) Diatomaceæ (Bacillariæ) Phæophyceæ (Melanophyceæ) Rhodophyceæ FUNGI Schizomycetes (Bacteria) Myxomycetes	Phycomycetes Ascomycetes (including yeasts) Basidionycetes Fungi Imperfecti ARCHIGONIATÆ Bryophyta (mosses) Lycopside (club mosses) Pteridophyta (ferns) Spermatophyta (seed plants)
Animals	
PROTOZOA Sarcodina Mastigophora Infusoria Sporozoa ROTIFERA CRUSTACEA	BRYOZOA (Polyzoa) PORIFERA (sponges) CÆLENTERATA (Hydrozoa) VERMES (worms) ARTHROPODA (insects, mollusks, etc.) VERTEBRATA (fish and amphibians)

Generic names may be taken from any source whatever, but like specific names usually give an indication of the appearance, history, origin, characters or properties of the species. For example, the common diatom *Asterionella formosa* is so designated because the radial arrangement of the cells forms a star-shaped (astral) body of pleasing form. If a species occurs in several varieties the variety may be indicated by a second adjective; in our example, *gracillima* meaning "most graceful." A further means of identification is provided by adding the name of the worker who first or best described the organisms; in our example, Heiberg.

Families and subdivisions of families are commonly designated by the name of one of the principal genera modified by a standard ending. Orders and sub-orders are named after one of the principal families modified by a different standard ending. The endings used by botanists and zoölogists are not the same; they are shown in the following

schedule. The zoölogical endings used are those of Calkins, 1926, and are not necessarily adhered to by all zoölogists.

ENDINGS USED BY BOTANISTS AND ZOÖLOGISTS TO DESIGNATE ORDERS,
FAMILIES AND TRIBES

	Botanical	Zoölogical
Order.....	ales	ida
Sub-order.....	ineæ	ina
Family.....	aceæ	idæ
Sub-family.....	oidææ	inæ
Tribe.....	ææ	same as for family or sub-family
Sub-tribe.....	inæ

Scope of Descriptions. — Of the many thousands of different species of microscopic organisms found in fresh water only a few are described in this book. These have been chosen chiefly because of their frequent occurrence and their important influence on the quality of the water in which they are found; in some instances they have been included as representative of a class or group in order that the attention of the student may be drawn to them. The reader is urged to extend his studies beyond the confines of the present volume.

The description of the organisms is not in many cases carried beyond the genus. To describe the different species belonging to the same genus would have been quite beyond the possibilities of a small work. The reader should remember, however, that under nearly all of the genera mentioned there are a number of common species. Similarly the plates do not include illustrations of all of the species commonly seen.

In the references given at the end of each chapter an attempt is made to list the reference works found most useful in identifying the organisms more definitely; a few of the less technical papers dealing with the morphology and biology of the various groups of organisms are also included.

Keys and Their Use. — Systematists have devised tabulations of the most conspicuous characters of the different groups to enable one to locate the name of an organism. Such tabulations are called keys and vary in usefulness according to their authors.

In the following chapters keys are provided wherever they will be of service in expediting the work of finding the name of the more common genera of fresh-water organisms. For example, suppose one to have a blue-green alga. On turning to the key on p. 456, one is confronted by a choice in No. 1, whether the organism is filamentous or not. On looking at the organism it is discovered to be filamentous. At the right

of the page under this category one sees the number 17. One turns to No. 17 in the key and is confronted by a choice of spores and heterocysts present or not. Supposing the organism is found to have heterocysts, the number at the right under this category, indicates that one should turn to No. 26. Under this number one has the choice of terminal hairs present or absent. Supposing the organism has no terminal hairs, one goes to No. 27 where the choice is filaments unbranched or branching. The organism is found to be unbranched when No. 28 is indicated. Under 28, assuming a sheath is conspicuous, with a single trichome in a sheath, one decides that the organism in question is *Microchæte*.

Such a key is known as a dichotomous key, since one has only a choice of two contrasting characters. For the most part, keys in this book are of this nature, but in some cases, to save space without sacrificing clearness, three or even more choices are given whenever the distinctions are clear cut.

Ecological Classification. — An ecological classification of aquatic organisms will be found in Chapter XXXII. Included in this classification are those organisms that have been observed to react in given ways to their environment. The list is intended for use more particularly in connection with studies of the pollution and natural purification of water. To this end many of the common genera found in relatively unpolluted water and not immediately responsive to recent or past pollution are omitted from the classification.

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CHAPTER XVII

ALGÆ

The term "algæ" is used to cover several phyla of primitive plants, capable of elaborating their own carbohydrate supply by means of photosynthesis, usually without highly differentiated vegetative structures. Most of them live entirely submerged in water, and several groups including the most highly developed in their vegetative structures are almost exclusively marine. They are found floating freely at the surface (plankton), attached to stones or clinging in gelatinous masses to the submerged portions of the more highly organized aquatic plants. A few prefer damp situations in which they do not become immersed at all, or only periodically become covered with water.

They are mainly distinguished from fungi by the presence of chlorophyll and consequently by their mode of life. Even in algæ that do not appear green to the observer, the chlorophyll is present but is masked by the presence of other pigments, as can easily be proved by the removal of the pigments in question. The method of reproduction varies very much in the different groups, from simple fission without even a well-developed nucleus as in the Cyanophyceæ to some of the most elaborate reproductive mechanisms known either in the animals or in other plant groups. As usual in biological classifications, the larger groups are separated on the basis of the reproductive mechanisms, but since these are usually accompanied by rather constant differences of color, we may recognize the groups on this basis.

Classification. — Six classes of algæ are commonly recognized.

1. **CYANOPHYCEÆ** (blue-green algæ or Myxophyceæ), usually blue-green in color but sometimes reddish or brownish, the pigment rather uniformly distributed in the cell. The stored product of assimilation is said to be glycogen. No well-defined nucleus present, reproduction by simple fission, cells of the colony or filament usually enclosed in a thick, gelatinous sheath, which, however, is very thin and inconspicuous in a few genera. Mostly fresh-water organisms.

2. **CHLOROPHYCEÆ** (green algæ), usually bright green in color, the pigment collected into a definite structure in the cytoplasm known as the chloroplast. The stored product of assimilation is starch. Nucleus present, reproduction usually by motile cells with two flagella of equal length except in the Conjugales which are frequently placed in a separate class on this account. Both marine and fresh-water representatives.

3. **XANTHOPHYCEÆ** (yellow-green algæ or Heterokontæ), usually yellowish in color, due to relatively large amounts of xanthophyll present. The stored product of

assimilation is a fatty substance. Nucleus present, reproduction by motile cells with two flagella of unequal length, the shorter inconspicuous and frequently not observed. Mostly fresh-water organisms.

4. DIATOMACEÆ (diatoms or Bacillariæ), usually brownish in color, the chlorophyll of the chloroplast masked by the brown pigment. The stored product of assimilation is a fatty substance. Nucleus present, asexual reproduction by fission, sexual reproduction by conjugation. Universal both in fresh and salt water.

5. PHÆOPHYCEÆ (brown algæ or Melanophyceæ), usually dark brown in color, the chlorophyll of the chloroplast masked by a brown pigment. The stored product of assimilation is starch. Nucleus present, reproduction by flagellated cells, at least the male cells. Mostly marine organisms. The unicellar organisms frequently classed with the Phæophyceæ are also classed with the protozoa by most American workers and are included in the latter group in the present work.

6. RHODOPHYCEÆ (red algæ), usually some shade of red, the chlorophyll of the chloroplast masked by a red pigment. The stored product of assimilation is starch. Nucleus present, the reproductive mechanism very elaborate with both male and female cells non-motile. Mostly marine organisms.

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CHAPTER XVIII

CYANOPHYCEÆ

The plants belonging to the Cyanophyceæ or Myxophyceæ are characterized by the presence of chlorophyll plus certain coloring substances known as cyanophyll, phycocyanine, phycoxanthine, etc., which are probably modifications of chlorophyll; by the absence of a nucleus and usually of starch grains; and by extremely simple but imperfectly understood methods of reproduction. The plants are one- or many-celled. By successive division of the cells they are commonly associated in families that take the form of filaments or of spherical or irregular masses.

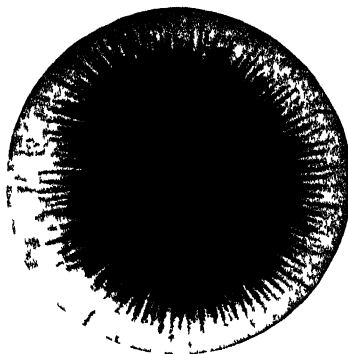
The cell wall is often distinct and sharply defined, but in some cases it is fused with a gelatinous mass in which the cells are embedded. This gelatinous matrix is more common in the terrestrial than in the aquatic species. The cell contents are usually granular and homogeneous.

The color varies considerably in different species and under different conditions. It is never a chlorophyll-green, but ranges from a color approaching that to a blue-green, orange-yellow, brown, red or violet. The coloring matter known as phycocyanine has a bluish color when viewed by transmitted light, and a reddish color when viewed by reflected light. This phenomenon is often observed in ponds where Cyanophyceæ are abundant. Looking directly at the pond, the water may have a reddish-brown color, while a bottle filled with the water and held to the light may present a decidedly bluish-green appearance. This is particularly true when the plants have begun to decay. The phycoxanthine is said to have a yellowish color. The liberation of gas bubbles from some species seems to have an effect on the color of the organisms. *Anabæna*, for example, may have a brownish-green color in a reservoir and a very light blue-green color after it has passed through the pipes of a distribution system, where the pressure has caused the gas to be expelled.

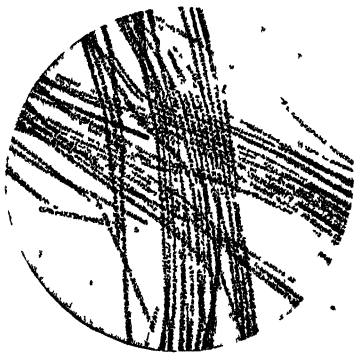
The Cyanophyceæ are usually separated into five or six groups, which are ranked by different writers as orders, families, or sections. The groups are here considered as families belonging to two orders.



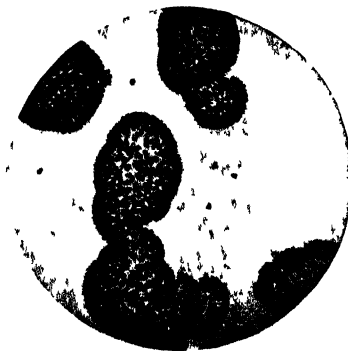
Anabaena



Rivularia



Aphanizomenon



Cœlosphærium.

PLATE D.

Photomicrographs of Typical Cyanophycææ.

KEY TO FRESH-WATER GENERA

1. Unicellular; cells often united loosely into families of various form but not into definite filaments, etc. 2
1. Cells united into simple or branching filaments. 17
2. Cells filiform, spirally twisted. *Spirulina*
2. Cells not filiform. 3
3. No reproduction other than by vegetative division of cells; no distinction of base and apex. 4
3. Reproduction by non-motile spores formed by division of contents of a cell, as well as by vegetative division; cells or families showing a distinction of base and apex. 16
4. Cells solitary or united in small indefinite families without general membrane 5
4. Cells united in more or less definite families, with general membrane. 6
5. Cells spherical. *Chroococcus*
5. Cells elongate, slender, fusiform. *Dactylococcopsis*
6. Families without definite form. 7
6. Families of definite form. 10
7. Membrane of successive generations persistent, the older enclosing the younger as distinct stratification. 8
7. Membrane diffuent; no stratification. 9
8. Stratum solid, formless; cells not in any order. *Gloeocapsa*
8. Stratum hollow; cells in fours. *Placoma*
9. Cells subspherical; division in three directions. *Aphanocapsa*
9. Cells elongate; division transverse. *Aphanothece*
10. Free floating. 11
10. Attached. *Oncobyrsa*
11. Cells not forming a single superficial layer to the family. 12
11. Cells forming a single layer. 15
12. Families spherical or subspherical. 13
12. Families cubical, alpine ponds. *Eucapsis*
13. Cells spherical or subspherical. 14
13. Cells elongate, mostly obpyramidal. *Gomphospheria*
14. Families clathrate. *Clathrocystis*
14. Families not clathrate. *Microcystis*
15. Family a hollow sphere. *Celosphaerium*
15. Family a membrane-like expansion. *Merismopedia*
16. Cells in short vertical series, laterally united into an extended encrusting stratum. *Radaisia*
16. Cells in short unbranched series. *Chamaesiphon*
17. No spores nor heterocysts. 18
17. Heterocysts present. 26
18. Filaments with no sheath, or exceptionally with a very evanescent one. 19
18. Filaments with a definite sheath. 20
19. Filaments regularly and uniformly spiral. *Arthrospira*
19. Filaments not at all or only slightly and irregularly spiral. *Oscillatoria*
20. Filaments each in a single sheath. 21
20. More than one filament in a sheath. 24
21. Filaments pseudo-branched. *Plectonema*
21. Filaments unbranched. 22
22. Sheaths mucous, diffuent, uniting the filaments into a submembranaceous stratum. *Phormidium*
22. Sheaths not mucous or diffuent, colorless or yellowish. 23
23. Tips of filaments uniting into erect penicillate tufts. *Symploca*
23. Filaments loose or matted but not in erect tufts. *Lyngbya*
24. Filaments scattered and relatively few in a sheath. *Hydrocoleus*
24. Many filaments forming a dense strand in the sheath. 25
25. Sheath firm, free. *Sirocoleus*
25. Sheath mucous, adherent. *Microcoleus*
26. No terminal hairs present. 27
26. Terminal hairs present. 41
27. Filaments unbranched. 28
27. Filaments with false or genuine branching. 34
28. Sheaths none or inconspicuous and diffuent. 29
28. Sheaths evident, trichome single in sheath. *Microchaete*
29. Filaments free, or adherent but in no definite form. 30

29. Filaments united in colonies of some definite form.	32
30. Heterocysts terminal.	<i>Cylindrospermum</i>
30. Heterocysts intercalary.	31
31. Cells discoid.	<i>Nodularia</i>
31. Cells globose to cylindrical.	<i>Anabaena</i>
32. Colony with firm epidermal layer.	<i>Nostoc</i>
32. No firm epidermal layer.	33
33. Colonies in form of minute, membranaceous scales.	<i>Aphanizomenon</i>
33. Colonies tubular.	<i>Wolleea</i>
34. False branching only; filaments of a single series of cells.	35
34. True branching; filaments often of several series of cells.	38
35. Trichome single in sheath.	36
35. Trichomes several in sheath.	37
36. Branch arising below a heterocyst, usually single	<i>Tolypothrix</i>
36. Branch arising between heterocysts; single or geminate.	<i>Scytonema</i>
37. Trichomes irregularly curved and twisted in sheath.	<i>Hydrocoryne</i>
37. Trichomes one or several in sheath, not curved; filaments in penicillate tufts.	<i>Desmonema</i>
38. Filaments free.	39
38. Filaments united into a definite stratum.	40
39. Filaments of a single series of cells.	<i>Hapalosiphon</i>
39. Filaments of more than one series of cells.	<i>Stigonema</i>
40. Stratum small, pulvinate, firm.	<i>Caposira</i>
40. Stratum expanded, rounded, soft	<i>Nostochopsis</i>
41. Filaments single or tufted, not in definite colonies.	42
41. Filaments definitely united.	44
42. Trichomes single in a sheath.	<i>Calothrix</i>
42. Trichomes more than one in a sheath.	43
43. Sheath including original trichome and branch.	<i>Dichothrix</i>
43. Sheath including many trichomes and branches.	<i>Polythrix</i>
44. Filaments erect, parallel, seldom branched, laterally united into a flat expansion.	<i>Isactis</i>
44. Filaments radiating, more or less branched.	<i>Rivularia</i> (incl. <i>Gloeotrichia</i>)

CLASSIFICATION AND DESCRIPTION

ORDER I. COCCOGONALES

Unicellular plants with spherical, oblong, or cylindrical cells enclosed in a tegument and associated in families that are surrounded by a universal tegument or immersed in a generally colorless, mucilaginous substance of varying consistency. Division takes place in one, two, or three directions, the cells after division usually remaining together forming an amorphous thallus. There are two families containing about thirty rather imperfectly defined genera.

FAMILY CHROOCOCCACEÆ. — Thallus mucous or gelatinous, amorphous, enclosing cells and families irregularly disposed.

Chroococcus. — Cells spherical, or more or less angular from compression, solitary or united in small families. Cell membrane thin or confluent in a more or less firm jelly. Cell contents pale bluish-green, rarely yellowish. Propagation by division in three directions. Several species are described. Most of them are terrestrial and not aquatic. The most common aquatic species are *C. turgidus*, the cells of which are from 10 to 25 μ in diameter, and *C. coherens*, the cells of which are from 3 to 6 μ in diameter. (Pl. IV, Fig. 5.)

Gloeocapsa. — Cells spherical, single or in groups; each cell surrounded by a vesiculiform tegument and groups of cells surrounded by an additional tegument. Cell membrane thick, lamellated, and sometimes colored. Division in three directions.

Cell contents bluish-green, brownish, or reddish. There are many described species, based on slight distinctions and variations in size and color. *Gloëocapsa* found in water usually has smaller cells and a more distinct tegument than *Chroococcus*. Comparatively few species are aquatic. (Pl. IV, Fig. 6.)

Aphanocapsa. — Cells spherical, with a thick, soft, colorless tegument, confluent in a homogeneous mucous stratum which is sometimes of a brownish color. Cell contents bluish-green, brownish, etc. The cells divide alternately in three directions. There are several species. The cells vary in size from 3 to 6 μ . (Pl. IV, Fig. 7.)

Microcystis. — Cells spherical, numerous, densely aggregated, enclosed in a very thin, globose mother-vesicle, forming solid families, singly or several surrounded by a universal tegument. Cell contents æruginous to yellowish-brown. The cells divide alternately in three directions. This genus represents a condition of frequent occurrence in the process of development of higher forms. There are several indistinct species common in water. The cells vary in size from 4 to 7 μ in diameter and the colonies from 10 to 100 μ . (Pl. IV, Fig. 8.)

Clathrocystis. — Cells very numerous, small, spherical or oval, æruginous, embedded in a colorless matrix. Multiplication by division of the cells within the thallus. The thallus is at first solid, then becomes saccate and clathrate (perforated); broken fragments are irregularly lobed. There is but one species — *C. æruginosa*. The cells are from 2 to 4 μ in diameter and the thallus from 25 μ to 5 mm. The species is widely distributed. (Pl. IV, Fig. 9.)

Celosphaerium. — Cells numerous, minute, globose or sub-globose, geminate, quaternate, or scattered, immersed in a mucous stratum. Cell contents æruginous, granulose. The thallus is globose, vesicular, hollow, the cells being found only on the outer surface. Multiplication takes place by division of the cells on the surface and by the escape and further development of certain peripheral cells. There is one common species, *C. kuetzingianum*. The cells are from 2 to 5 μ in diameter and the thallus from 50 to 500 μ . (Pl. IV, Fig. 10.)

Merismopedia. — Cells globose or oblong, æruginous or brownish, with confluent teguments. Division in two directions. The thallus is tabular, quadrate, free-swimming, the cells being arranged in groups of 4, 8, 16, 32, 64, 128, etc. There are several indistinct species. The diameter of the cells varies from 3 to 7 μ . (Pl. IV, Fig. 11.)

FAMILY CHAMÆSIPHONACEÆ. — Plants often showing a difference between apical and basal regions, solitary or associated in families or colonies, usually epiphytic or attached to shells; reproduction by cell division, by division of filaments into fragments, or by means of non-motile gonidia formed by the division of the contents of a mother cell or gonidangium.

ORDER II. HORMOGONALES

Multicellular plants, the cells of which dividing in one direction, form filaments, often enclosed in a tubular sheath. The filaments (trichomes) may be either simple or branched. Reproduction occurs by means of hormogones or resting gonidia.

FAMILY NOSTOCACEÆ. — Plants composed of rounded cells loosely united into filaments, or trichomes, and sometimes embedded in jelly. The filaments do not branch and never terminate in a hair point. They sometimes form large masses. There are three kinds of cells — ordinary vegetative cells, joints, or articles; hetero-

cysts; and spores. The ordinary cells are spherical, elongated, or compressed. The cell contents are bluish-green or brownish, and are usually granular. The heterocysts are cells found at intervals in the filaments. They are spherical, elliptical, or elongated, and are usually somewhat larger than the vegetative cells. Their cell contents are generally clear or very finely granular, and usually of a light bluish-green color. The cell wall is sharply defined, and there are two polar lumps of gelatinous material that cause them to adhere to the adjoining cells. The function of the heterocysts is unknown, but they are thought to be in some way connected with the process of reproduction. The spores are usually much larger than the vegetative cells. They are spherical, elliptical, or cylindrical. Their cell contents are usually very granular and dark-colored. They seem to be more highly differentiated than the contents of the vegetative cells. The spores are heavy, and will sink in water when freed from the filaments. Multiplication takes place by division of the vegetative cells, by means of the spores, and by means of hormogonia, or internal parts of the filaments which separate from them and form new plants. The character and position of the heterocysts and spores form the chief basis for the division of the Nostocaceæ into genera. The classification is very indefinite.

Nostoc. — Cells globose or elliptical; heterocysts usually globose and somewhat larger than the vegetative cells; spores oval and but little larger than the heterocysts. Spores and heterocysts are both intercalated in the filaments, rarely terminal. The filaments are enclosed in a gelatinous envelope, and are flexuously curved and irregularly interwoven. They often form gelatinous fronds or thalli surrounded by a firm membrane. The thalli vary in diameter and are sometimes of great size. There are many species, both terrestrial and semi-aquatic. The true Nostoc is seldom found in drinking water. (Pl. IV, Fig. 12.)

Anabaena. — Vegetative cells spherical, elliptical, or compressed in a quadrate form. Heterocysts much larger than the vegetative cells, subspherical, elliptical, or barrel-shaped, of a pale yellowish-green color, and intercalated in the filament. Spores globose or oblong-cylindrical, equal to or somewhat larger than the heterocysts, rarely smaller. The filaments are moniliform; are without sheaths; are straight, curved, circinate, or intertwined; have a bluish-green or brownish color; and are often free-floating. There are several important but imperfectly defined species. The most common species are *A. flos-aquæ* and *A. circinalis*. The vegetative cells of the former are from 5 to 7 μ in diameter; those of the latter are from 8 to 12 μ . (Pl. IV, Figs. 13 and 14.; Pl. V, Fig. 1.)

Cylindrospermum. — Vegetative cells globose, elliptical, or compressed, homogeneous or granular. Heterocysts terminal, spherical, or oval, but little larger than the cells. Spores adjacent to the heterocysts, oval or cylindrical, much larger than the cells. The filaments are moniliform, sheathless, and sometimes taper slightly. There are few species, and these resemble some forms of *Anabaena* and *Sphærozyga*. (Pl. V, Fig. 2.)

Aphanizomenon. — Vegetative cells cylindrical, closely connected, granular, and with little color. Heterocysts rare, intercalated, oval, but little larger in diameter than the cells. Spores very rare, intercalated, not adjacent to heterocysts, cylindrical, with rounded ends, sometimes of dark olive color. The filaments are cylindrical, slightly tapering, and densely agglutinated in fascicles, occasionally free. The fascicles are often of considerable size. Diameter of filaments 4 to 6 μ . This genus is sometimes mistaken for *Oscillatoria* or *Anabaena*. (Pl. V, Fig. 3.)

FAMILY OSCILLATORIACEÆ. — Filaments without heterocysts or spores, with or without sheath, not terminating in a hair point, single or associated in bundles

enclosed in a common sheath. The division of the filaments into cylindrical cells is indistinct. Multiplication is said to take place by hormogons, i.e., parts of the trichomes which separate from the rest of the filament.

Oscillatoria. — Cells shortly cylindrical, disk-shaped in end view, closely united into a simple, branchless, sheathless filament. The filaments are straight or somewhat curved, occasionally fasciculate, and have rounded ends. The color is bright bluish-green, steel-blue, etc. The filaments when in active vegetative state possess characteristic spontaneous oscillating movements. There is a large number of species, that vary in diameter from 1 to 50 μ , and have cells differing in shape and in color. There are but few free-floating forms. (Pl. V, Fig. 4.)

Lyngbya. — Filaments enclosed singly in a sheath, branchless, but with occasional appearance of branching during multiplication, sometimes combined to form a membranaceous stratum. Cells united into short trichomes, with rounded ends, not continuous in the sheath, but separated by clear spaces. Cell contents blue-green, granular. Sheaths pellucid, hyaline. Propagation is said to take place by hormogonia and by gonidia. There are many species, terrestrial and aquatic. (Pl. V, Fig. 5.)

Microcoleus. — Filaments rigid, articulate, crowded together in bundles, enclosed in a common mucous sheath, either open or closed at the apex. Sheath ample, colorless, rarely indistinct. Several species, chiefly terrestrial. (Pl. V, Fig. 6.)

FAMILY SCYTONEMATACEÆ. — Filaments with lateral ramifications (false branching) in which some of the cells change into heterocysts; enclosed in a sheath. The cells divide transversely. The ramifications are produced by the deviation of the trichome and emergence through the sheath. The branches do not have a hair point. There are several genera.

Scytonema. — Sheath enclosing a single trichome, composed of subspherical or subcylindrical cells, with scattered heterocysts. Color bluish or yellowish-green. Ramification takes place by a folding of the trichomes, followed by rupture of the sheath and the emergence of one or two portions of the folded trichome at right angles to the original filament. These branched filaments produce interwoven mats. Multiplication is said to take place by microgonidia. There are many species, terrestrial and aquatic. The plant is not found free-floating. (Pl. V, Fig. 7.)

FAMILY STIGONEMATACEÆ. — Trichomes enclosed in an ample sheath, profusely branched. Branches are formed by longitudinal division of certain cells so as to form two sister cells, the inferior of which remains a part of the trichome, while the other, by repeated division, grows into a branch. The filaments often contain 3, 4 or more series of cells. Propagation is said to take place by means of microgonidia.

Stigonema. — Cells one-, two-, or many-seriate, in consequence of their lateral division or multiplication. The cells have a distinct membrane and the sheaths are large. The plant is never found free-floating. (Pl. V, Fig. 8.)

FAMILY RIVULARIACEÆ. — Filaments free or agglutinated into a definite thallus, terminating at the apex in a hair-like extremity. Heterocysts usually basal. Trichomes articulated like *Oscillatoria*, parallel or radially disposed. Spores, when present, cylindrical, generally adjacent to the basal heterocyst.

Rivularia. — Filaments radial, agglutinated by a firm mucilage, and forming well-defined hemispherical or bladderly forms. Heterocysts basal. No spores formed. Ramifications produced by transverse division of the trichomes. Color greenish to brownish. Sheaths usually distinct. Several species, terrestrial and aquatic. Occasionally found free-floating. (Pl. V, Fig. 9.)

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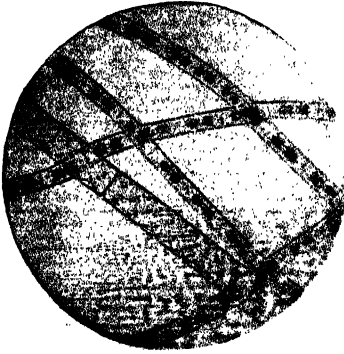
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CHAPTER XIX

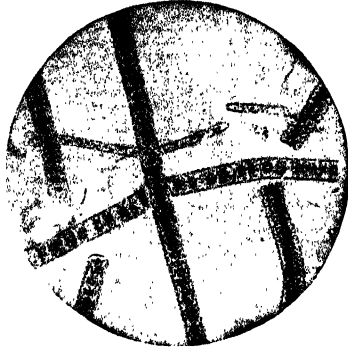
CHLOROPHYCEÆ

The plants belonging to the Chlorophyceæ are characterized by the presence of true chlorophyll, a nucleus, starch grains, and often by a cell wall made of cellulose. They are "algæ" in the strictest sense of the term. They cover a great range of complexity. Some of them are minute, unicellular forms scarcely distinguishable from the cyanophyceæ; others as the Volvocales resemble the protozoa; while others as the Charales are large, branching, multicellular forms, doubtfully included among the algæ, and very similar to plants much higher in the scale of life. Most of them are aquatic, but a few are terrestrial. Their color is almost always a bright chlorophyll-green, but occasionally it is yellowish-brown or even a bright red.

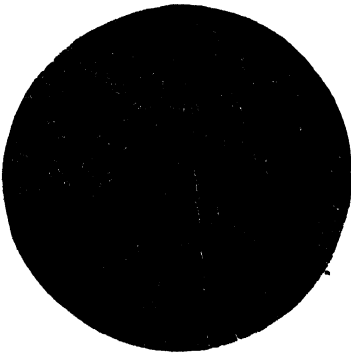
Reproduction. — The Chlorophyceæ increase by the ordinary processes of cell division observed in the higher forms of plant life. The cells may separate after division, or they may remain associated in colonies or in simple or branching filaments. Reproduction takes place either asexually, i.e., without the aid of fecundation, or sexually. There is but one general method of asexual reproduction, namely, the formation within the cell of spores, which become scattered and give rise to new cells. There are three general types of sexual reproduction. The simplest is the formation in the cells of zoöspores, which become liberated and ultimately copulate with other zoöspores. Two of these zoöspores become attached by their ciliated ends, their contents become fused, and a zygospore results. After a period of rest the zygospore may develop into a new plant, or may break up into other spores. The second type of sexual reproduction is known as conjugation. Two cells come in contact, and by means of openings in the cell walls their contents become fused. A zygospore (sometimes two) is formed, which, after a period of rest, gives rise to new plants. The highest form of sexual reproduction takes place by the formation of a rather large female oöspore which becomes fertilized by small male cells or sperms. This mode of reproduction is analogous to that observed in the higher plants. Many of the Chlorophyceæ exhibit the phenomenon of "alternation of generations," by which is meant the continued propagation of the plants by asexual processes with the occasional intervention of the sexual processes.



Spirogyra and *Zygnema*.



Zygnema.



Draparnaldia.



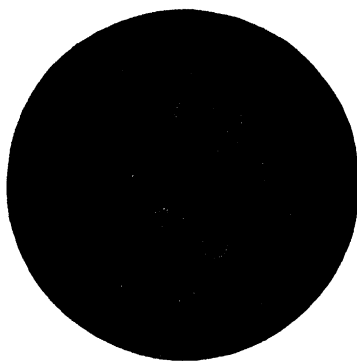
Batrachospermum.

PLATE E.

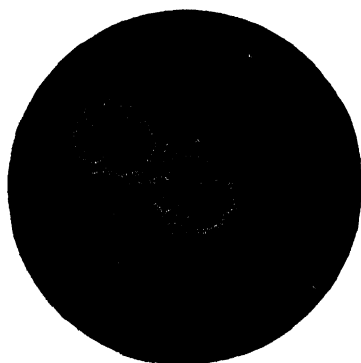
Photomicrographs of Typical Chlorophyceæ.



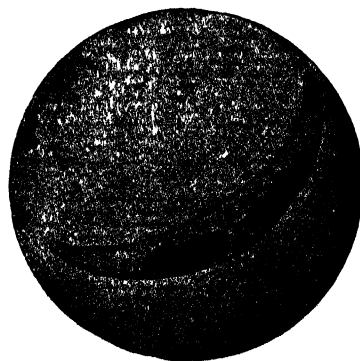
Pedastrum.



Tetraspora.



Cosmarium.



Closterium.

PLATE F.

Photomicrographs of Typical Chlorophyceæ.

KEY TO FRESH-WATER GENERA

1. Cells solitary or in filaments, divided into two symmetrical halves, generally with a median constriction. 94
1. Cells very large, in highly developed structures, branches in whorls at definite nodes, reproductive mechanisms very highly developed. 123
1. Cells otherwise. 2
2. Cells in ordinary condition with motile cilia. 3
2. Cells without cilia, or ciliate only in reproduction. 15
3. Cells solitary. 4
3. Cells united in families or colonies. 7
4. Cells fusiform. **Chlorogonium** 5
4. Cells spherical to ovoid. **Hematococcus** 5
5. Protoplasmic threads passing through the cell wall. 6
5. No such threads. **Chlamydomonas** 6
6. Chromatophore grass-green. **Cryptoglena**
6. Chromatophore blue-green. **Spondylomorom** 8
7. Colony without gelatinous sheath. **Pyramimonas** 9
7. Colony with general gelatinous sheath. **Gonium** 10
8. Cells without membrane, obconical with wide longitudinal ribs. 11
8. Cells with membrane. **Pandorina** 11
9. Cells arranged in a plate, with cilia on one side. 12
9. Cells arranged otherwise. 13
10. Cells conical, crowded, apices approximately near the center of the colony. 13
10. Otherwise. 13
11. Colony with cells uniform in size. **Eudorina** 13
11. Colony with small vegetative and large reproductive cells. **Platydorina** 13
12. Colony ellipsoidal or spherical, poles not differentiated. **Volvox** 14
12. Colony flattened, horseshoe-shaped, poles differentiated. 14
13. Sexual reproduction by antheridia and oögonia; colonies usually of over 500 cells. **Pleodorina** 16
13. No sexual reproduction known; colonies usually of less than 300 cells. **Besseyosphaera** 16
14. Vegetative and reproductive cells in separate parts of the colony. 37
14. Vegetative and reproductive cells intermingled. **Characium** 17
15. Cells solitary or temporarily and loosely adherent. 18
15. Cells more or less permanently united. 18
16. Attached by a stipe or rhizoids. **Trochiscia** 19
16. Without organs of attachment. 21
17. Cells branching. 21
17. Cells spherical, covered with ridges or short spines. **Vaucheria** 21
17. Cells angular, often with projections. **Dichotomosiphon** 22
17. Cells rounded, cylindrical or filiform. **Tetraëdron** 23
18. Branching lateral. 23
18. Branching dichotomous. 23
19. Cells with distinct central polyedral mass. **Cerasterias** 27
19. Rays from common center without distinct central mass. **Thamniastrum** 27
20. Rays unbranched. 27
20. Rays dichotomously divided. 27
21. Cell with one or more projections. **Dicranochaete** 23
21. Cell without projections. 23
22. Cell bearing a branching hair. **Micractinium** 24
22. Cell bearing spines or papillae. 24
23. Cell wall with thick gelatinous sheath. 25
23. Cell wall without gelatinous sheath. 25
24. Cell thin-walled with several spines. **Centrosphaera** 26
24. Cell thick-walled with rounded or conical papilla. **Acanthosphaera** 26
25. Spines relatively thick in lower half, extremely slender in upper half. 26
25. Spines of uniform diameter, or gradually tapering, but without sharp distinction between upper and lower halves. **Francela** 28
26. Cell dividing longitudinally only. **Lagerheimia** 28
26. Cell dividing in three directions. 28
27. Cells spherical. 28
37. Cells not spherical. 34

28. Without pyrenoid, chloroplast usually red.	29
28. With pyrenoid.	30
29. Wall of uniform thickness, chromatophores disk-shaped.	<i>Palmellococcus</i>
29. Wall becoming thicker at one side, ultimately forming a stipe-like prolongation.	<i>Urococcus</i>
30. Wall thin, homogeneous.	31
30. Wall thick, protoplast usually red.	<i>Hamatococcus</i>
31. Chromatophores numerous, disk-shaped.	32
31. Chromatophores single.	33
32. Chromatophores small, rounded.	<i>Eremosphera</i>
32. Chromatophores larger, angular.	<i>Excentrosphera</i>
33. Zoospores 4 to 8 in a cell.	<i>Chlorosarcina</i>
33. Zoospores many in a cell.	<i>Chlorococcum</i>
34. Cell reniform.	<i>Nephrocytium</i>
34. Cell ovoid or broadly fusiform.	<i>Oöcystis</i>
34. Cell fusiform or aciculate.	35
34. Cell cylindrical.	<i>Stichococcus</i>
35. Cell dividing by two planes at right angles, daughter cells maintaining a definite order, enclosed loosely in gelatin.	<i>Quadrigula</i>
35. Cells dividing obliquely.	36
36. Cells 4 to 8 μ diam., 225 to 530 μ long, many-seriate pyrenoids.	<i>Closteropsis</i>
36. Cells proportionately shorter, one or more pyrenoids.	<i>Ankistrodeasmus</i>
37. Cells in definite order, united by common gelatin.	38
37. Cells definitely arranged.	43
38. Gelatin amorphous.	39
38. Gelatin of definite form.	41
39. Cells ovoid to short-fusiform.	<i>Coccomyxa</i>
39. Cells spherical or subspherical.	40
40. Broken pieces of cell wall persisting in gelatin.	<i>Schizochlamys</i>
40. Wall entirely dissolving in gelatin.	<i>Palmella</i>
41. Gelatin forming a membranous expansion or tube.	<i>Tetraspora</i>
41. Gelatinous sheath fusiform.	<i>Fusula</i>
41. Gelatin forming a branching filiform thallus.	42
42. Cells large, distantly seriate.	<i>Hormotila</i>
42. Cells small, uniformly distributed.	<i>Palmodictyon</i>
43. Cells included in general gelatinous sheath	44
43. Cells without general gelatinous sheath	56
44. Gelatinous thallus filiform.	45
44. Gelatinous thallus a horizontal layer of densely-packed vertical filaments.	48
44. Gelatinous thallus spherical, tubercular or branching.	<i>Chætophora</i>
44. Gelatinous thallus pyriform, attached.	<i>Apiocystis</i>
44. Gelatinous thallus rounded, free.	49
44. Gelatinous thallus of indefinite form.	50
45. Gelatinous thallus unbranched.	46
45. Gelatinous thallus branched.	47
46. Gelatinous sheath transversely lamellate.	<i>Radioflum</i>
46. Gelatinous sheath not transversely lamellate.	<i>Geminella</i>
47. Cells fusiform.	<i>Elakatothrix</i>
47. Cells spherical, walls persisting after cell division.	<i>Palmodictyon</i>
48. Akinetes and zoospores in intercalary sporangia.	<i>Chlorotylum</i>
48. Akinetes and aplanospores terminal; zoospores unknown.	<i>Wittrockiella</i>
49. Colony of 2 to 8 regularly placed cells, usually surrounded by dark bands	<i>Gloiotenium</i>
49. Colony otherwise.	<i>Gliococcus</i>
50. Cell wall persistent after cell division.	51
50. Cell wall not persistent after cell division.	52
51. Cells spherical.	<i>Gliocystis</i>
51. Cells elongate.	<i>Dactylothece</i>
51. Cells crescent-shaped.	<i>Kirchneriella</i>
52. Cells in twos and fours, with bristles; chromatophore blue-green.	<i>Glaucocyste</i>
52. Cells without bristles; chromatophore grass-green.	53
53. Cells in one plane.	54
53. Cells not in one plane.	<i>Gliocystopsis</i>
54. Cells without prominences.	55
54. Cells with projections from the edges.	<i>Tetrastrum</i>

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|--|------------------------|
| 55. Cells spherical, in groups of four; groups without definite order in colonies. | Westella |
| 55. Cells of various form, usually flattened, never spherical; groups of four usually forming aggregates in one plane. | Crucigenia |
| 56. Daughter cells persistent in wall of mother cell; no other union. | 57 |
| 56. Cells united to form a disk. | 59 |
| 56. Cells elongate, united side to side | 60 |
| 56. Cells arranged otherwise. | 61 |
| 57. Cells ovoid. | 58 |
| 57. Cells reniform. | Nephrocystium |
| 58. Chromatophore blue-green. | Glaucocystis |
| 58. Chromatophore grass-green. | Oöcystis |
| 59. Cells trapezoidal, two to a colony. | Euastropsis |
| 59. Cells of various form, many in a colony | Pediastrum |
| 60. In one plane. | Scenedesmus |
| 60. In two planes. | Tetradasmus |
| 61. Cells arranged symmetrically about a center | 62 |
| 61. Cells forming an expanded membrane, flat or tubular. | 64 |
| 61. Cells forming filaments. | 73 |
| 62. Cells lunate or sickle-shaped. | Selenastrum |
| 62. Cells not lunate or sickle-shaped. | 63 |
| 63. Colony in the form of a depressed sphere; cells in rings. | Phytomorula |
| 63. Cells attached by one end at center of colony; radiating in all directions. | Actinastrum |
| 63. Cells attached at center of colony by stipes. | 64 |
| 64. Stipes short unbranched. | 65 |
| 64. Stipes branched. | 66 |
| 65. Cells with spinous projections. | Sorastrum |
| 65. Cells without spinous projections. | Colastrum |
| 66. Cells in series on gelatinous strands. | Dictyocystis |
| 66. Cells at ends of strands. | 67 |
| 67. Cells spherical. | Dictyosphaerium |
| 67. Cells reniform, ovoid or cordate. | Dimorphococcus |
| 68. Membrane attached to substratum by one surface. | 69 |
| 68. Attached only at base. | 71 |
| 69. Cells with hairs or bristles. | 70 |
| 69. Cells without hairs or bristles. | Protoderma |
| 70. Bristles with sheathed base. | Coleochaete |
| 70. Hairs without sheathed base. | Chaetopeltis |
| 71. Flat. | 72 |
| 71. Tubular. | Enteromorpha |
| 72. Chromatophore stellate; no motile spores. | Prasiola |
| 72. Chromatophore discoid; motile spores produced. | Monostroma |
| 73. Filaments forming a net. | Hydrodictyon |
| 73. Filaments partly of more than one series of cells. | 74 |
| 73. Filaments forming a single series of cells. | 75 |
| 74. Cells ultimately dividing to form a plane. | Schizogonium |
| 74. Cells ultimately forming a parenchymatous mass. | Schizomeris |
| 75. Filaments unbranched. | 76 |
| 75. Filaments branched. | 84 |
| 76. Division of cell leaving rings about the filament at upper end of cell. | Oögonium |
| 76. No such rings. | 77 |
| 77. Protoplast ovoid, fusiform, dividing to form a pair of conical cells, base to base with a common lamellate wall. | Cylindrocapsa |
| 77. Otherwise. | 78 |
| 78. Chromatophore net-shaped or breaking up into small disks. | 79 |
| 78. Chromatophore continuous, parietal. | 80 |
| 78. Chromatophore an axillary plate. | 82 |
| 78. Chromatophore one or more spiral bands. | 83 |
| 78. Chromatophores two rounded, excentric disks. | Pleurodiscus |
| 78. Chromatophores two, stellate. | Zygnema |
| 78. Chromatophores in zones; reproduction by oöspores and antheridia. | Sphaeroplea |
| 79. Regularly cylindrical, attached. | Chaetomorpha |
| 79. More or less irregular, not attached. | Rhizoclonium |
| 80. Chromatophore a disk or plate. | Stichococcus |

80. Chromatophore a sonate band.	81
81. Apical and basal cells attenuate.	Uronema
81. Apical and basal cells little attenuated.	Ulothrix
82. Spore ovoid with three longitudinal ridges and many fine cross lines.	Debarya
82. Spore lentiform or with outline more or less angled, smooth or scrobiculate.	Mougeotia
83. Vegetative cells conjugating directly.	Spirogyra
83. Short conjugating cells cut off from long vegetative cells.	Temnogyra
84. With hairs or setæ.	85
84. Without hairs or setæ.	80
85. Setæ sheathed at base.	Chætospheridium
85. No sheaths.	86
86. Setæ bulbous at base.	87
86. Setæ not bulbous at base.	88
87. Frond prostrate.	Herpoteiron
87. Frond erect.	Bulbochaete
88. Cells of main filaments of different appearance from those of the definitely outlined branch system.	Draparnaldia
88. Axes or successive orders differing only in size.	Stigeoclonium
89. Branches few-celled, rhizoidal.	Rhizoclonium
89. Branches similar to axes.	90
90. Erect filaments densely packed, forming a definite stratum.	91
90. Otherwise.	92
91. Sporangia terminal.	Gongrosira
91. Sporangia intercalary.	Leptosira
92. Very minute, no cross walls at base of branch.	Microthamnion
92. Larger, cross wall usually at base of branch.	93
93. Reproduction by large cylindrical, conical or barrel-shaped akinetes.	Pithophora
93. Reproduction by minute zoospores or zoogametes.	Cladophora
94. Cells solitary, separating after division.	95
94. Cells continuing united.	110
95. No constriction or only a very slight one between two semi-cells.	96
95. Semi-cells separated by a distinct constriction.	100
96. Cells straight.	97
96. Cells more or less distinctly crescent or bow-shaped.	99
97. Chromatophore parietal, spiral.	Spirotenia
97. Chromatophore a single plate in each cell.	Mesotenium
97. Chromatophore one or more in each semi-cell.	98
98. Chromatophore stellate.	Cylindrocystis
98. Chromatophore with entire edges.	Penium
98. Chromatophore with notched edges.	Netrium
99. Curvature slight; chromatophore with convex tip, quite filling cell.	Roya
99. Curvature slight or marked; chromatophore with concavity at tip, leaving space for apical vacuole.	Closterium
100. Semi-cells several times as long as broad; median constriction slight.	101
100. Semi-cells not much longer than broad; median constriction usually marked.	105
101. Semi-cell with a notch at apex.	102
101. Semi-cell without notch at apex.	104
102. Cell wall with rings of furcate processes.	Triploceras
102. Cell wall smooth.	103
103. Apical notch wide, apical angles with spines.	Ichthyocercus
103. Apical notch narrow, apical angles rounded.	Tetmemorus
104. Bases of semi-cells plicate.	Docidium
104. Bases of semi-cells not plicate.	Pleurotenium
105. Cells in vertical view 3- or more-angled or with radiate projections; rarely fusiform.	Staurostrum
105. Cells in vertical view oval to fusiform, elongate at right angles to line of front view.	106
106. Cells with spines.	107
106. Cells without spines.	108
107. Semi-cell with central protuberance.	Xanthidium
107. No central protuberance.	Arthrodesmus
108. Semi-cell without deep incision.	Cosmarium
108. Semi-cell with distinct, often quite deep incision.	109
109. Semi-cells more or less lobed and incised, of various outline, usually with one or more hemispherical prominences near base.	Euastrum

109. Cells strongly compressed, of circular or oval outline, semi-cells usually deeply and sharply lobed and incised, usually without conspicuous prominences.	Microsterias	
110. Cells united by branching gelatinous bands, not in simple filaments.		111
110. Cells united into simple filaments.		112
111. Bands slender, colonies loose.	Cosmocladium	
111. Bands stout, colonies compact.	Oëcardium	
112. Protoplasm colored purple.	Ancyclonema	
112. Protoplasm not colored purple.		113
113. Cells cylindrical, length many times the diameter; outer wall with spines or roughness.		114
113. Cells of various form, not long cylindrical.		115
114. Chromatophore axial.	Gonatozygon	
114. Chromatophore parietal, spirally twisted.	Genicularia	
115. Cells united by apical processes.		116
115. Cells united directly, without processes.		119
116. Cells squarish in front view, much compressed, united in ribbon-like series.	Microsterias foliaceae	
116. Cells not much compressed nor squared in front view.		117
117. Apical processes short, meeting similar processes from next cell.	Sphaerosozma	
117. Apical processes longer.		118
118. Each semi-cell with two processes, overlapping on next cell, but not meeting processes of the latter.	Onychonema	
118. Each cell united to the next cell by three cylindrical processes.	Streptonema	
119. Median constriction slight.		120
119. Median constriction marked.		121
120. Filaments surrounded by a wide gelatinous sheath.	Hyalotheca	
120. Filaments without gelatinous sheath.	Desmidium	
121. Cells angular or with radiate arms in vertical view.		122
121. Cells elliptical in vertical view.	Spondylosium	
121. Cells circular with two small opposite prominences in vertical view.	Gymnozyga	
122. Cells angular in vertical view, semi-cells symmetrical.	Desmidium, pp.	
122. Cells quadrate with prolonged angles; prolongations laterally asymmetrical in vertical view; semi-cells asymmetrical in front view.	Phymatodocis	
123. Sporangium with coronula of 5 unicellular rays.		124
123. Sporangium with coronula of 5 biocellular rays.		127
124. Diccious; no stipular ring to whorls of leaves.	Tolytellopsis	
124. Stipular ring present.		125
125. Diccious.	Chara, pp.	
125. Monœcious.		126
126. Sporangium below the antheridium.	Lamprothamnus	
126. Sporangium laterally between antheridia.	Lynchnothamnus	
126. Sporangium above the antheridium.	Chara, pp.	
127. Antheridium terminal on a leaf, or on a leaflet not of the last order.	Nitella	
127. Antheridium on a unicellular lateral leaflet.	Tolytella	

CLASSIFICATION AND DESCRIPTION

SUBCLASS I. ISOKONTEÆ

Cells solitary or in colonies and showing the same great range of variation in shape, organization of the colony, interior structure, and method of reproduction noted in the description of the class.

ORDER I. VOLVOCALES

Cells normally ciliate and motile during the vegetative phases of the life cycle. Solitary or organized into colonies of definite form that generally contain a definite number of cells. Protoplasts rarely naked, usually with a differentiated outer cellulose covering that may be partially gelatinized. Shape of protoplast spherical, ovoid, disciform,

pyramidate or irregularly radiate. Cells with a single cup-shaped to irregular chloroplast with or without pyrenoids; with 2 to 4 cilia at the anterior end; generally with a disciform to bacilliform eyespot, two to several contractile vacuoles and one nucleus. *Palmella* stages frequently arising in the unicellular members. Asexual reproduction by division of cell contents into 2 to 16 zoöspores; or a development of special or all cells of the colony into autocolonies. Sexual reproduction by isogamous zoögametes, heterogamous zoögametes or small motile sperms and large non-motile oöospheres.

FAMILY CHLAMYDOMONADACEÆ. — Cells normally solitary and motile during the vegetative phases of the life history. Spherical, ovoid, cylindrical, sub-acicular, compressed or quadrately projected. Chloroplast cup-shaped to laminate, lateral or posterior in position, with or without pyrenoids. Cells uninucleate, with or without an eyespot, with two cilia of equal length that generally have two contractile vacuoles at their base. Cells at times becoming immobile and developing into amorphous colonies enclosed by a wide gelatinous envelope. Asexual reproduction by the division of cell contents to form 2 to 16 biciliate zoöspores. Sexual reproduction by the fusion of isogamous zoögametes. *Chlamydomonas* is the principal genus. It is listed as a protozoön in this work (see page 514).

FAMILY VOLVOCACEÆ. — Cells always motile and in cœnobias of definite shape that contain a definite or an indefinite number of cells. Colonies enclosed in a homogeneous, hyaline, gelatinous sheath. Cells of colony all alike or differentiated into vegetative and reproductive cells. Vegetative cells spherical, ellipsoid, pyriform or disciform; with or without connecting cytoplasmic processes; always biciliate. Chloroplasts generally cup-shaped and containing one pyrenoid. Vegetative cells usually with an eyespot and two contractile vacuoles. Asexual reproduction by the division of all or certain of the cells in the colony to form autocolonies. Sexual reproduction by a division of all cells in the colony to form isogamous zoögametes or heterogamous zoögametes of slightly different size; or the formation of sperms and oöospheres. Zygotes with smooth or sculptured walls.

Gonium. — Colonies of 4-8-16 cells arranged in a flat quadrangular plate and embedded in a common gelatinous matrix or connected by broad gelatinous strands. Cells ovoid to pyriform. Four- and eight-celled colonies with the cilia on the same side; sixteen-celled colonies with the four central cells having their cilia on the same side and the twelve marginal cells with radially arranged cilia. Asexual reproduction by simultaneous division of all cells in the colony to form autocolonies or by the formation of 2 to 4 zoöspores in each cell. Sexual reproduction isogamous by a fusion of biciliate zoögametes. There is but one species, *G. pectorale* Mueller. (Pl. VII, Fig. 6.)

Pandorina. — Colonies always motile, spherical to subspherical, containing 4-8-16-32 cells mutually compressed to form a hollow sphere with a small open space in the center. Colony enclosed by a fairly copious, firm, hyaline, homogeneous, gelatinous sheath. Cells pyriform to angular by mutual compression, with pointed ends toward the center of the colony; biciliate at the flattened distal ends, the two cilia lying close together while passing through the colonial sheath and then becoming quite divergent. Asexual reproduction by simultaneous division of all cells of the colony to form autocolonies that are liberated by a gelatinization of the envelope. Sexual reproduction by a division of each cell of the colony into 16 to 32 zoögametes,

which show indications of heterogamy in the slight difference in size and motility of the pairs that fuse to form the smooth-walled zygote. *P. morum* is the only common species. (Pl. VII, Fig. 5.)

Eudorina. — Colonies always motile, spherical or slightly elongate of 16–32–64 cells lying some distance from one another and arranged to form a hollow sphere near the periphery of the homogeneous, hyaline, gelatinous sheath. Cells spherical with or without a beak at the point of origin of the two cilia, which are parallel while passing through the colonial sheath and then divergent. Sexual reproduction heterogamous, dioecious, with all the cells of a colony developing into large immobile oöospheres or plate-like masses of 32 to 64 fusiform sperms; or monœcious with four cells forming sperms and the rest oöospheres. Zygote smooth-walled. *E. elegans* is a common species. (Pl. VII, Fig. 4.)

Volvox. — Colonies always motile, spherical to ovoid, containing a large number of cells arranged in a single layer just within the periphery of the homogeneous, hyaline, gelatinous colonial sheath. Cells differentiated into those for vegetative purposes, asexual and sexual reproduction. Vegetative cells close together or some distance from one another, with or without cytoplasmic connections of varying thickness. Cell-shape spherical, ovoid, or disciform. Number of cells in a colony varying from 200 to 22,000. Asexual reproductive cells few in number (rarely more than 20) and forming autocolonies by repeated division, the young colonies migrating to the center of the cornobium at a certain stage of their development. Sexual reproduction heterogamous, monœcious or dioecious, with a development of 6 to 400 cells into spherical oöospheres and few or many of the cells into antheridia that contain 16 to 256 biciliate fusiform sperms. Zygote with a smooth or stellate wall. Colonies sometimes reach a millimeter or more in diameter. (Pl. VII, Fig. 3.)

ORDER II. PROTOCOCCALES

Vegetative cells non-motile, solitary or in colonies. Colonies amorphous and frequently embedded in a gelatinous sheath; or of definite shape and with or without a gelatinous sheath. Colonies capable or incapable of increasing the number of their cells after they are formed. Cells variously shaped, generally with one chloroplast and pyrenoid. Asexual reproduction by zoöspores, fragmentation of the colony, autospores or autocolonies. Resting akinetes known in some species. Sexual reproduction by isogamous zoögametes known but not found in a majority of the genera.

FAMILY PALMELLACEÆ. — Cells spherical, ovoid or reniform; rarely solitary, generally united to form colonies which are at times of macroscopic size. Colonies embedded in a gelatinous matrix, capable of an increase in size by the vegetative division of the constituent cells. Chloroplasts generally single, cup- to disk-shaped and parietal; or star-shaped and central; with or without pyrenoids. Reproduction by the fragmentation of the colony or by zoöspores. Sexual reproduction known in certain genera.

Gloeocystis. — Cells spherical; solitary or in small colonies of 8 cells or less. Each cell and the whole colony surrounded by a hyaline, lamellated, gelatinous sheath. Asexual reproduction by a fragmentation of the colony through the softening of the gelatinous envelope or by zoöspores. Akinetes known. The size of the cells varies

from 2 to 12 μ in diameter and the colonies from 10 to 100 μ . Color green, sometimes reddish, the gelatinous envelope hyaline to ochraceous. (Pl. V, Fig. 10.)

Tetraspora. — Colonies macroscopic or microscopic; attached or free-floating; gelatinous, spherical, cylindrical, expanded or variously lobed. Cells spherical, generally in groups of four toward the periphery of the homogeneous, hyaline, gelatinous colonial sheath. Chloroplast cup-shaped, parietal; sometimes diffuse, with one pyrenoid. Each cell with two or four long, hyaline gelatinous bristles (pseudocilia). Asexual reproduction by the direct metamorphosis of vegetative cells into biciliate zoöspores. Sexual reproduction by a division of vegetative cells to form 4 to 8 biciliate zoögametes. Cells from 3 to 12 μ in diameter. (Pl. V, Fig. 12.)

Palmella. — Cells spherical ovoid or oblong, surrounded by a thick confluent sheath, forming an amorphous colony by alternate division of the cells in all directions. Size of the cells varies 1 to 15 μ , colonies often very large, color usually green. (Pl. V, Fig. 11.)

FAMILY DICTYOSPHERIACEÆ. — Cells in spherical or ovoid colonies, the number in young colonies a multiple of two, in older colonies indefinite. Colonies with or without a gelatinous sheath. Cells held together in fours or eights by the persistence of the old mother-cell wall in repeatedly branching four-armed thongs or irregular threads. Chloroplast single, cup-shaped or filling the entire cell. Reproduction by the division of cells into autospores which remain attached to the mother colony until accidentally broken away.

Dictyosphaerium. — Cells spherical, ovoid or reniform, connected with one another by cruciately or dichotomously branching threads. Colony enclosed by a hyaline, homogeneous gelatinous envelope that is spherical or ovoid in shape. Reproduction by the division of the cell into 2 or 4 daughter cells which remain connected to the colony by the old mother-cell wall that divides into 2 or 4 parts to form the branching thread system connecting the cells. Resting akinetes known. (Pl. VI, Fig. 3.)

Dimorphococcus. — Cells in groups of four and held in an irregular free floating colony by the branching remains of the old mother-cell wall, not enclosed by a gelatinous sheath. Each colony of 4 cells lying in one plane with 2 cells ovoid to cylindrical with rounded ends, and 2 cells reniform to cardioid. Reproduction by the division of any cell into 4 daughter cells which remain attached to the colony by the threadlike remains of the old cell wall until accidentally broken away. *D. lunatus* A. Braun is a common species. (Pl. VI, Fig. 5.)

FAMILY PROTOCOCCACEÆ. — Cells solitary and spherical; or in colonies containing an indefinite number of cells that are spherical and mutually compressed or in very short, irregular filaments. Cell wall delicate to heavy, smooth or variously sculptured. Chloroplast single, parietal and disciform with or without pyrenoids. Cells capable of vegetative division. Reproduction, aside from vegetative division of cells, by zoöspores or aplanospores. *Protococcus viridis* is a common species. (Pl. VI, Fig. 6.)

FAMILY AUTOSPORACEÆ. — Cells solitary or in colonies that are generally of a definite shape and without a gelatinous sheath. Cells variously shaped; with a single chloroplast and pyrenoid as a rule. Cells incapable of vegetative division to form two similar daughter cells. Reproduction by division of the cell contents into 2, 4, 8, 16, 32 or 64 cells which assume the shape of the mother cell before their liberation (autospores). In the colonial species the autospores from any one cell are organized to form the new colony (autocolony) before their liberation. Motile asexual reproductive cells or sexual reproduction unknown.

Oöcystis. — Cells ovoid, ellipsoid or cylindrical with rounded to somewhat pointed ends, generally symmetrical but never curved. Cell wall smooth, without spines, frequently with nodular thickenings at the poles. Chloroplasts one to many, parietal, disciform, stellate or reticulate, with or without pyrenoids. Cells solitary or in temporary colonies of 2, 4, 8, 16 or more enclosed by a partially gelatinized and greatly swollen old mother-cell wall. Reproduction by autospores, with often 3 to 4 cell generations enclosed by the same wall.

Nephrocytium. — Cells ovoid, reniform or oblong-elliptic; generally in colonies of 2, 4, 8, or 16 cells within the partially gelatinized remains of the old mother-cell wall. Arrangement of cells in young colonies spiral, in old colonies irregular. Chloroplast single and expanded at first, later fragmenting and becoming diffuse; pyrenoid single. Reproduction by the formation of 2, 4, 8 or 16 autospores in each cell. (Pl. VI, Fig. 4.)

Tetraëdron. — Cells solitary, free-floating, flattened or isodiametric, triangular, quadrangular or polyangular. Angles simple or produced into simple or furcate spines. Cell walls smooth or verrucose. Chloroplasts one to many, parietal, disciform to angular; or completely filling the cell; with or without pyrenoids. Reproduction by division of cell contents into 4 or 8 autospores that are liberated by the rupture of the old mother-cell wall. (Pl. VI, Fig. 7.)

Ankistrodesmus. — Cells acicular to fusiform; straight, lunate or sigmoid; ends of cells gradually tapering to a point; solitary or loosely aggregated without order in temporary colonies that are not embedded in a gelatinous envelope. Chloroplast single, parietal, sometimes fragmented into small pieces; with or without a pyrenoid. Reproduction by the formation of several autospores in any cell. (Pl. VI, Fig. 2.)

Crucigenia. — Cœnobe of four cells quadrately arranged with a quadrangular open space at the center and frequently connected to the other cœnobias by a gelatinous envelope or the remains of the old mother-cell walls. Multiple cœnobias forming a flat plate one cell in thickness containing 4 to 64 cells. Cells flattened; ovoid, triangular, trapezoidal or semicircular in front view; with a smooth cell wall. Chloroplasts 1 to 4 parietal, disciform to laminate with or without pyrenoids. Reproduction by autocolony formation. (Pl. VII, Fig. 2.)

Scenedesmus. — Colony a flat (rarely curved) plate of ellipsoidal, ovoid, or acicular cells with cell number always a multiple of two. Cells in lateral contact and in one or two rows. Cell wall smooth, corrugated, granulate, or spicate; with or without marginal or lateral teeth or spines. Chloroplast single, parietal, and laminate in young cells; frequently filling entire cell in older colonies; pyrenoids single, central or eccentric. Reproduction by autocolonies from any or all cells of the colony, the number of cells in the colony not necessarily the same as that of the mother colony. (Pl. VI, Fig. 8.)

Cœlastrum. — Colony a hollow sphere of 2, 4, 8, 16, 32, 64 or 128 cells. Cells spherical, ovoid or pyramidal; compressed or with large intercellular spaces; enclosed in a very delicate gelatinous sheath. Daughter colonies sometimes remaining joined in multiple colonies by the remains of the old mother-cell wall. Reproduction by autocolony formation in any cell of the old colony. (Pl. VII, Fig. 1.)

Sorastrum. — Cells pyriform, semilunar, or reniform; united to form a spherical colony of 8, 16, 32, 64 or 128 cells. Center of colony a polyhedral body from which gelatinous strands radiate to each cell. Distal side of cells with 1, 2 or 4 spines. Chloroplast diffuse, with one pyrenoid. The reproduction is unknown; presumably

by autocolonies as in *Cœlastrum*, possibly by zoöspores as in *Pediastrum*. (Pl. VI, Fig. 12.)

FAMILY HYDRODICTYACEÆ. — Cells cylindrical or flattened and hexagonal to trapezoidal with 1, 2 or 4 projections. Colonies of definite shape; cylindrical with a large number of cells; or disciform with 2, 4, 8, 16, 32, 64 or 128 cells. Cells incapable of division after their formation. Chloroplast single, parietal, laminate to reticulate, frequently filling the entire mature cell; with one to many pyrenoids. Asexual reproduction by the division of contents of any cell to form zoöspores that swarm within the old cell wall or within a gelatinous vesicle extruded from the cell. Zoöspores becoming apposed in the shape of the adult cell when they come to rest. Sexual reproduction by isogamous zoögametes which fuse in pairs to form angular resting cells which form colonies on their germination.

Hydrodictyon. — Cells macroscopic, oblong-cylindrical, with rounded ends; united to form a free-floating, reticulate, saccate cœnobium. Meshes of nets 3- to 12-sided (generally 5- or 6-sided). Cells cœnocyctic; chloroplast parietal, reticulate at first, later diffuse; pyrenoids single in very young cells, several hundred in mature cells. Asexual reproduction by the division of cell contents into very many biciliate zoöspores which swarm about in and become arranged as in the mature cœnobium within the old mother-cell wall. Sexual reproduction by zoögametes which are similar in structure to zoöspores but smaller, the zoögametes fusing in pairs to form a spherical zygote after their liberation through a pore in the cell wall. Zygote germinating after a short period of rest into 2 to 5 large uni- or bi-ciliate zoöspores that form irregular polyhedral cells on coming to rest. Polyhedral cells ultimately germinating into 200 to 300 zoöspores that are liberated in a vesicle and form a net as in asexual reproduction. (Pl. VI, Fig. 9.)

Pediastrum. — Cœnobium disciform to stellate, free-floating of 2, 4, 8, 16, 32, 64 or 128 cells arranged in a layer one cell in thickness. Cœnobium entire, perforate or clathrate. Marginal cells polygonal, with 1, 2 or 4 processes that sometimes terminate in a tuft of long hyaline setæ. Interior cells polygonal, without processes. Cell wall smooth, granulate or covered with a meshwork of fine ridges; without a gelatinous sheath. Chloroplasts parietal and disciform at first, later filling the entire cell; with 1 to 4 pyrenoids. Cells cœnocyctic. Asexual reproduction by the division of contents of any cell into 2, 4, 8, 16, 32, 64 or 128 biciliate zoöspores that are extruded in a gelatinous vesicle and, after a short period of swarming, on becoming quiescent appose themselves in the same position as the mature cells of the cœnobium. A fusion of biciliate zoögametes into polyhedral resting cells has been observed. On germination these zygospores form 8, 16 or 32 zoöspores that behave as in asexual reproduction. There are many species; perhaps *P. boryanum* and *P. duplex* are the most common. (Pl. VI, Fig. 11.)

ORDER III. SIPHONALES

Cœnocyctic, unicellular plants when in the vegetative state; cells tubular or utricle-shaped, often branched. Cell contents green, granular. Propagation by sexual fertilization, asexual zoöspores, or by microgonidia.

FAMILY VAUCHERIACEÆ. — Plants consisting of elongated, robust tubular filaments, more or less branched, growing in tufts. Chlorophyll granules are evenly distributed on the inside walls of the cells, and starch grains and oil globules are

conspicuous. Sexual propagation takes place by means of oöspores fertilized by sperms. The oögonia are lateral, sessile, or borne on a simple pedicel; the antheridia usually develop on the same filament. Asexual propagation takes place by means of zoöspores produced in a terminal sporangium. The zoöspores are ciliated, but go through a resting period before germinating. Propagation also takes place by means of microgonidia produced in the vegetative cells.

Vaucheria. — The characteristics are described under the family. There are many species, aquatic and terrestrial. (Pl. IX, Fig. 7.)

ORDER IV. SIPHONOCLODIALES

Cœnocytic multicellular plants, usually consisting of much branched filaments, chloroplasts usually parietal, often reticulate, occasionally in simple plates. Reproduction by the union of similar ciliated gametes. Asexual reproduction by zoöspores and aplanospores. Sexual reproduction isogamous by means of zoögametes; or heterogamous by large non-motile oöospheres and small sperms. The order is largely marine; only one genus, *Cladophora*, is frequently found in fresh water.

FAMILY CLADOPHORACEÆ. — Filaments simple or branched, with septations between the cells complete. Walls of cells generally heavy, homogeneous or lamellose, without a gelatinous sheath. Cells with numerous parietal disciform chloroplasts or a single reticulate chloroplast; with a single large central vacuole. Asexual reproduction by zoöspores and akinetes. Sexual reproduction by means of zoögametes. *Cladophora* is the only important fresh water genus. (Pl. IX, Fig. 9.)

ORDER V. ULOTRICHALES

Cells usually in a simple or branched thallus rarely forming disciform thalli one cell in thickness. Thallus generally attached, rarely free-floating; naked or enclosed in a tough and leathery or copious and gelatinous, homogeneous or lamellated sheath. Cells generally with a single parietal laminate to cup-shaped chloroplast with one or more pyrenoids. Cell shape usually cylindrical with flattened ends, rarely cylindrical with rounded ends, spherical or cubical. Asexual reproduction by vegetative cell division of cell contents to form bi- or tetraciliate zoögametes; or heterogamous by the fusion of tetraciliate zoögametes of different size or the union of large non-motile oöospheres and small biciliate sperms.

FAMILY ULOTRICHACEÆ. — Simple filaments enclosed at times by a copious hyaline homogeneous or radially fibrillar, gelatinous sheath. Cell-walls delicate or thick, homogeneous or lamellated. Cells containing a nucleus and one parietal laminate or cup-shaped chloroplast with one or more pyrenoids. Asexual reproduction by vegetative cell division, aplanospores or biciliate zoöspores. Sexual reproduction isogamous by a fusion of bi- or tetraciliate zoögametes. *Ulothrix* is one of the larger genera of fresh water. (Pl. IX, Fig. 10.)

FAMILY CHÆTOPHORACEÆ. — Branched filaments, or discoidal or parenchymatous thalli. Branches of thalli generally attenuate at apices and sometimes ending in long hair-like processes. Thallus naked or enclosed in a copious hyaline, homogeneous gelatinous sheath. Chloroplast single, parietal and laminate to girdle-shaped; with one or more pyrenoids, nucleus single. Asexual reproduction by aplanospores and bi- or tetraciliate zoöspores. Sexual reproduction isogamous by a fusion of biciliate zoögametes.

Chætophora. — Filaments arising from a palmelloid base, and united by a firm gelatinous substance into thalli of definite form; filaments repeatedly branched, of about the same diameter throughout, tips of branches often in fascicles, frequently terminating in long setæ. Chloroplast a parietal band with one or more pyrenoids. The plants are attached to sticks, stones, etc., and are common in clear running water in spring, less common in quiet water. (Pl. X, Fig. 3.)

Stigeoclonium. — Branching filaments, terminal cells pointed or prolonged into a seta; chloroplast a parietal band, filling the smaller cells, zonate in the larger. Asexual reproduction by tetraciliate zoöspores with a red eyespot; also by akinetes which produce biciliate zoöspores, by aplanospores and also by a palmella stage. Sexual reproduction by conjugation of biciliate gametes with a red eyespot. (Pl. X, Fig. 2.)

Draparnaldia. — Filaments united by a soft, gelatinous coating, not forming a thallus of definite form; main filaments attached by basal rhizoids, more or less branched, stout, bearing dense lateral fascicles of small branches, much smaller than the main filaments, often setiferous. Chloroplasts in the stem and larger branches a parietal band, sometimes perforated, with numerous pyrenoids; in the cells of the small branches, a layer covering the wall, with few pyrenoids. Asexual reproduction only from cells of the small branches, by tetraciliate zoöspores with red eyespot, germinating immediately; also akinetes and aplanospores; sexual reproduction by conjugation of tetraciliate gametes, which however may germinate without copulation. Common plants of running water, chiefly in spring; distinguished from *Stigeoclonium* and *Chætophora* by the sharp contrast between the main stems and the small branches; also from *Chætophora* by the thin, amorphous character of the gelatinous coating. (Pl. X, Fig. 1.)

SUBCLASS II. AKONTEÆ

Cells solitary or in filaments, variously shaped with one or more grass-green chloroplasts that usually contain more than one pyrenoid. Asexual reproduction ordinarily by vegetative cell division, rarely by aplanospores, never by zoöspores. Sexual reproduction isogamous, always by aplanogametes, never by ciliate zoögametes. The lack of cilia in every type of reproduction is the great characteristic of the division. It includes but one order, the Conjugales, which forms a very homogeneous though diverse group.

ORDER I. CONJUGALES

Unicellular or multicellular plants. The multicellular forms have no terminal vegetation and are destitute of true branches. The chlorophyll masses are arranged in plates, bands, or stellate masses. Starch

grains are abundant. Multiplication by division in one direction. Reproduction by zygospores resulting from copulation and conjugation of two cells, or by azygospores formed without copulation. There are two families that are very different in their general characteristics, but that agree in their mode of reproduction.

FAMILY DESMIDIACEÆ. — The Desmids form a large, well-defined group of unicellular algae. They are characterized by an apparent division of the cell into two symmetrical halves. The cells are of various sizes and forms, often curious or ornamental, single or joined together forming a filament. The transverse constriction is sometimes deep, sometimes slight, and occasionally absent. The cell wall is firm, almost horny. Some writers have imagined that it was slightly silicified. The cell is surrounded by a mucous covering and sometimes by a layer of gelatin. The cell contents are green and granular. Starch grains are numerous. At the ends of some of the cells there are clear spaces in which are seen granules that occasionally have a vibratory movement. Cyclosis, or a circulation of granules in the watery fluid next to the cell wall, may be observed in some species. Some species of desmids exhibit voluntary movements of the entire cell. *Closterium*, for example, shows certain oscillations and backward- and forward-gliding movements, supposed to be due to the secretion of threads of mucus. Multiplication takes place by cell division and by conjugation. In the first case the two halves of the cell stretch apart and become separated by a transverse partition; new halves ultimately form on each of the original halves, so that two symmetrical cells result. These afterward separate. (See Pl. VIII, Fig. A.) Sexual propagation by conjugation takes place as follows: Two cells approach and each sends out a tube from its center. These tubes meet, swell hemispherically, and, by the disappearance of the separating wall, become united into a rounded zygospore with a thick tegument and sometimes with bristling projections. This zygospore, after a period of rest, loses its contents through a rent in the wall, and a new cell is formed which ultimately becomes constricted and assumes the shape of the parent cell. (See Pl. VIII, Figs. B to F.)

Some of the common genera are described below. The enormous number of species makes a detailed analysis impracticable.

Penium. — Cells straight, cylindrical or fusiform, not incised nor constricted in the middle; ends rounded. Chloroplasts axillary; containing starch granules. Cell membrane smooth, finely granulated, or longitudinally striated. Individuals free-swimming. (Pl. VII, Fig. 7.)

Closterium. — Cells simple, elongated, lunate or crescent-shaped, entire, not constricted at the center. Cell wall thin, smooth or somewhat striated. The chloroplasts are generally arranged in longitudinal laminae, interrupted in the middle by a pale transverse band. At each end there is a clear, colorless, or yellowish vacuole in which minute "dancing granules" may be seen. (Pl. VII, Figs. 8 to 10.)

Docidium. — Cells straight, cylindrical or fusiform, elongated, constricted at the middle. The semi-cells are somewhat inflated at the base and are often separated by a suture. Ends rounded, truncated or divided. Transverse section circular. The chloroplast has a parietal or axillary arrangement. Terminal vacuoles with "dancing granules" are observed in some species. (Pl. VII, Fig. 11.)

Cosmarium. — Cells oblong, cylindrical, elliptical, or orbicular, with margins smooth, without spines; not deeply constricted; ends rounded or truncate and entire; end view oblong or oval. Chloroplasts parietal or concentrated in the center of the semi-cells. Cell walls smooth, punctate, warty, or rarely spinous.

The zygospore is spherical, tuberculated or spinous. (Pl. VII, Fig. 12, and Pl. VIII, Figs. A to F.)

Tetmemorus. — Cells cylindrical or fusiform, slightly constricted in the middle, narrowly incised at each end, but otherwise entire. Cell wall smooth. (Pl. VII, Fig. 13.)

Xanthidium. — Cells single or geminately concatenate, inflated, very deeply constricted; semi-cells compressed, entire, spinous, protruding in the center as a rounded, truncate, or denticulate tubercle. Cell wall firm, armed with simple or divided spines. The zygospores are globose, smooth or spinous. (Pl. VIII, Figs. 1 and 2.)

Arthrodesmus. — Cells simple, compressed, deeply constricted, semi-cells broader than long, with a single spine on each side, but otherwise smooth and entire. (Pl. VIII, Fig. 3.)

Euastrum. — Cells oblong or elliptical, deeply constricted; semi-cells emarginate and usually incised at their ends; sides symmetrically sinuate or lobed, provided with circular inflated protuberances; viewed from the vertex, elliptical. The zygospores are spherical, tuberculose or spinous. (Pl. VIII, Fig. 4.)

Micrasterias. — Cells simple, lenticular, deeply constricted; viewed from front, orbicular or broadly elliptical; viewed from the vertex, fusiform, with acute ends; semi-cells three- or five-lobed; lateral lobes entire or incised; end lobes sinuate or emarginate and sometimes with angles bifid or produced. (Pl. VIII, Fig. 5.)

Staurostrum. — Cells somewhat similar to those of *Cosmarium* in front view, but angular in end view; angles obtuse, acute, or drawn out into horn-like processes. Cell wall smooth, punctate or granular, hairy, spinulose, or extended into arms or hair-like processes. Chloroplasts concentrated at the center of the semi-cells, with radiating margins. The zygospores are spined. (Pl. VIII, Figs. 6 and 7.)

Hyalotheca. — Cells short, cylindrical, usually with a slight obtuse constriction in the middle; circular in end view. The cells are closely united into long filaments, enclosed in an ample, colorless gelatinous sheath. The chloroplast, in end view, has a radiate appearance. (Pl. IX, Fig. 1.)

Desmidium. — Cells oblong-tabulate, somewhat incised; in end view, triangular or quadrangular; united into somewhat fragile filaments. Chloroplasts in each semi-cell concentrated and radiate to the angles. Zygospores smooth, globose or oblong. (Pl. IX, Fig. 2.)

Sphærozosma. — Cells bi-lobed, elliptical, or compressed, deeply incised, forming filaments which are almost moniliform or pinnatifid, surrounded by a gelatinous sheath. Chloroplasts somewhat radiate. (Pl. IX, Fig. 3.)

FAMILY ZYGNEACEÆ. — Multicellular plants, composed of cylindrical cells joined into filaments. Cell wall lamellose. Chloroplasts are stellate, axillary laminae, or spiral bands. Starch grains, etc., conspicuous. Propagation by zygospores resulting from copulation, which takes place by the union of two filaments. The filaments come into proximity, the cells put out short processes, which unite, forming tubular passages between pairs of cells. Through these connecting tubes the cell contents of one cell pass into and unite with the cell contents of another. This results in the formation of a zygospore often clothed with a triple membrane. Copulation is said to be scalariform when opposite cells of two filaments unite by ladder-like tubes, geniculate when the cells become bent and unite at the angles, and lateral when the process takes place between two adjoining cells of the same filament. The family is sometimes divided into two tribes, the *Zygnemeæ* and *Mesocarpeæ*. In the second tribe the spore formed is not a true zygospore. It is formed by a flowing together of

only a part of the cell contents. The zygospores germinate by putting forth a single germ tube, which elongates by transverse division into a filament.

Spirogyra. — Cells cylindrical, sometimes replicate, or folded in at the ends. Chlorophyll arranged in one or several parietal spiral bands winding to the right. Copulation scalariform, sometimes lateral. Zygospores always within the wall of one of the united cells. There are very many species, differing in size of cells, number and arrangement of spirals, replication at the end of cells, character of the zygospore, etc. (Pl. IX, Figs. 4 and 5.)

Zygnema. — Cells with two, axillary, many-rayed chloroplasts near the central cell nucleus, containing one or more starch granules. Copulation scalariform or lateral. Zygospore in one of the united cells. (Pl. IX, Fig. 6.)

Mougeotia. — Cells cylindrical, generally several times as long as broad. Chloroplast single, forming an axial plate extending the whole length of the cell and generally with several pyrenoids. Conjugation generally scalariform with the zygote formed in the conjugation tube.

SUBCLASS III. STEPHANOKONTÆ

Cells in simple or branched filaments, attached, cylindrical, the terminal cells setiform in the genus *Bulbochæte*, rounded in *Ædogonium*, cells with one nucleus, with ribbon-like chloroplasts having several pyrenoids. Cell division is very characteristic in this subclass. The cell when about to divide develops a ring in the subapical portion of the cell wall which rapidly enlarges by intercalary growth, the wall of the mother cell ruptures just outside the ring, and the ring expands to a cylindrical shape for the new cell wall. This leaves characteristic ridges at the end of the cell which serve to identify this group immediately. Asexual reproduction by large zoöspores, which bear a crown of cilia at the anterior end. Sexual reproduction by oöospheres and motile sperms that also bear a crown of cilia. Plants monœcious or diœcious; when diœcious the male plants are either dwarf, i.e., produced in short filaments which usually develop epiphytic on the female plant, or much larger and independent. There is but a single family, the *Ædogoniaceæ* with three well known genera, *Ædogonium*, *Bulbochæte* and *Ædo-cladium*.

CHARACEÆ

The Characeæ are plants that occupy an intermediate position between the algæ and the higher plants. Each plant consists of an assemblage of long tubular cells, having a distinct central axis, with whorls of branches projecting at regular intervals at points called "nodes." The branches are sometimes spoken of as leaves, but they are quite similar to the stem. At the lower end of the stem some of the branches (rhizoids) are root-like and serve to give attachment and

stability to the plant. Reproduction takes place by a peculiar sexual process. Oöspheres or archegones form at the base of the branches and are fertilized by peculiar antherozoids found near them.

There are two common genera, *Nitella* and *Chara*. In *Nitella* the stems and branches are simple and naked; the leaves are in whorls of 5 to 8 and without stipules; the leaflets are large and often many-celled; the sporocarps arise singly or in clusters in the forkings of the leaves, and each has a crown of two superimposed whorls of five cells each. In *Chara* the stems and lower branches are usually corticated, i.e., there is a central tube surrounded by smaller tubes, sometimes spirally arranged, forming a cortex; the leaves are in whorls of 6 to 12, and usually with one or two stipules; the leaflets are always one-celled; the sporocarps arise from the upper side of the leaves, and each has a crown of one whorl of five cells. These plants exhibit beautifully the phenomenon of cyclosis, or circulation of protoplasm. Some species of *Chara* secrete calcium carbonate, and from this arises their popular name, "stone-worts." (Pl. XIX, Fig. 8.)

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CHAPTER XX

XANTHOPHYCEÆ

The cells in this class vary from microscopic to macroscopic in size, and are free-floating, sessile or epiphytic. The cell shape varies from spherical to ovoid, pyriform, cylindrical or irregular. Cells solitary, in colonies of regular or irregular shape, or united to form simple filaments. Colonies or individual cells with or without a gelatinous sheath. Chromatophores one to many, parietal, generally disciform, rarely laminate, yellowish-green in color and without a pyrenoid. The product of assimilation is oil, never starch. Cells may be either uninucleate or multinucleate. The asexual reproduction may be by the formation of zoöspores or by vegetative cell division. The zoöspores are ovoid to pyriform, with two cilia of unequal length, with the short cilium so short and closely appressed to the cell that it appears to be uniciliate. The sexual reproduction insofar as known is isogamous by zoögametes that are similar in structure to the zoöspores.

The various genera of the Xanthophyceæ or Heterokontæ were formerly scattered throughout the different families of the chlorophyceæ. They were first segregated as a subclass parallel to the Isokontæ, etc., of the Chlorophyceæ; hence the name Heterokontæ. They were finally raised to the distinction of a separate class on account of the fundamental differences in metabolism and phylogenetic relationships as shown by the zoöspores.

KEY TO FRESH-WATER GENERA

- | | |
|--|---------------------------|
| 1. Vegetative cells motile. | 2 |
| 1. Vegetative cells non-motile. | 5 |
| 2. Several chromatophores present, often amœboid. | Chloramœba |
| 2. Two parietal chromatophores. | 3 |
| 3. Cell with anterior portion fixed, posterior portion amœboid. | Chloromonas |
| 3. Cell not as above. | 4 |
| 4. Cell ovoid, amœboid, sometimes wholly pseudopodial. | Heterochloris |
| 4. Cell lens-shaped, laterally compressed, chromatophores on the broad sides. | Phakomonas |
| 5. Cells in colonies surrounded by a gelatinous sheath. | 6 |
| 5. Cells without a gelatinous sheath. | 9 |
| 6. Colony amorphous. | 7 |
| 6. Colony arborescent. | Mischococcus |
| 7. Surface of gelatinous sheath warty, groups of four cells in each wart. | Chlorosaccus |
| 7. Surface otherwise. | 8 |
| 8. Gelatinous sheath, pyriform, usually attached, cells regular with several chromatophores. | Leeuwenia |
| 8. Cells arranged radiately in small groups, two chromatophores. | Dictyosphaeriopsis |
| 9. Cells solitary or colonial, not filamentous. | 10 |

9. Cells arranged in filaments.	24
10. Cells solitary.	11
10. Cells arranged in colonies.	23
11. Cells spherical to slightly ellipsoidal.	12
11. Cells otherwise.	16
12. Cells sessile on a long slender stipe.	<i>Peroniella</i>
12. Cells free-floating.	13
13. Cells over 20 μ in diameter.	<i>Botrydiopsis</i>
13. Cells mostly under 10 μ in diameter.	14
14. Cells not strictly spherical, especially when arranged in short filaments.	<i>Heterococcus</i>
14. Cells spherical.	15
15. Cells with thin cell wall, one chromatophore and swarm spores.	<i>Pleurochloris</i>
15. Cells with thick wall and several chromatophores, swarm spores unknown.	<i>Chlorobotrys</i>
16. Cells sessile.	17
16. Cells free-floating.	19
17. Cells multinucleate, long cylindric.	<i>Ophiocytium</i>
17. Cells uninucleate.	18
18. Reproduction by direct formation of swarm spores.	<i>Characiopsis</i>
18. Reproduction not by direct formation of swarm spores.	<i>Chlorothecium</i>
19. Without spines or appendages.	20
19. With spines or warts.	22
20. Cells fusiform.	<i>Chlorocloster</i>
20. Cells ovoid with one end tapering to a point.	<i>Monodus</i>
20. Cells cylindric.	21
21. Uninucleate, thin-walled.	<i>Bumilleriopsis</i>
22. Multinucleate, thick-walled.	<i>Ophiocytium</i>
22. Cells short cylindric, apposed along their sides, almost quadrate with a spine at each corner.	<i>Pseudotetraëdron</i>
22. Cells elongate, uninucleate, with a spine on each rounded end.	<i>Centrictactus</i>
23. Colonies of long cylindrical or spiral cells, arborescent, or attached to each other by stalks in a more or less radial manner.	<i>Ophiocytium</i>
23. Colonies in a gelatinous envelope.	<i>Botryococcus</i>
24. Cell walls show in optical section definite H-shaped thickenings involving the ends and portions of the side wall, filaments usually breaking between these thickenings rather than between cells.	25
24. Cell walls not as above.	<i>Heterococcus</i>
25. All cells alike in the filament.	<i>Tribonema</i>
25. Between the H-shaped thickenings are thin-walled cells	<i>Bumilleria</i>

CLASSIFICATION AND DESCRIPTION

ORDER HETEROCOCCALES

Cells spherical, ovoid, saccate, or elongate-cylindrical; solitary or in colonies of definite or indefinite shape, free-floating or sessile. Cells of the colonies embedded in a hyaline, gelatinous sheath or a tough elastic membrane. Chromatophores one to many, yellowish-green to nearly grass-green in color and with oil, not starch, the assimilation product. Asexual reproduction by vegetative cell division and formation of zoöspores with cilia of unequal length. Sexual reproduction by a fusion of zoögametes that are similar in structure to the zoöspore is known in certain genera.

FAMILY BOTRYOCOCCACEÆ. — Cells always in free-floating colonies of definite or indefinite shape, number of cells in colony generally indefinite. Colony enclosed by a sheath that may be hyaline or colored, gelatinous or leathery. Cells usually ovoid, rarely spherical; with 1 to 2 parietal laminate to disciform, yellowish-green to nearly

grass-green chloroplasts. Asexual reproduction by vegetative cell division. Zoöspore formation or sexual reproduction unknown. The principal genus is *Botryococcus*. (Pl. VI, Fig. 1.)

FAMILY OPHIOCYTTACEÆ. — Cells sessile or free-floating, solitary or colonial. Length of cells generally several times the breadth. Cells straight, curved, sigmoid or spiral, with or without terminal spines. Chromatophores yellowish-green, annular, parietal, and few in number; or disciform parietal and numerous; asexual reproduction by division of cell contents to form 4 to 16 aplanospores or 2 to 8 biciliate zoöspores with cilia of unequal length. Sexual reproduction unknown. *Ophiocytium* is the best known genus. (Pl. VI, Fig. 10.)

ORDER HETEROTRICHALES

Cells in simple, unbranched filaments, with or without a gelatinous sheath. Cell wall firm, fairly thick, lamellated in structure and breaking down into H-shaped pieces at the time of reproduction. Chromatophores few to several, yellowish-green disciform and parietal, cells usually uninucleate. Asexual reproduction by the formation of aplanospores or by zoöspores with two cilia of unequal length. Sexual reproduction by the fusion of isogamous zoögametes. There is but one family in the order, whose characters are the same as those of the order. *Tribonema*, or *Conferva* of some of the older treatments, is the best known genus. (Pl. IX, Fig. 8.)

CHAPTER XXI

DIATOMACEÆ

The Diatomaceæ or Bacillariæ comprise a group of minute vegetable forms whose exact position in the scale of life has been the subject of much controversy. The early writers considered them to belong to the animal kingdom because of the power of movement that some of them possess. Later they became recognized as plants and were classified as a class of *algæ*. They differ from other unicellular organisms in the possession of siliceous cell walls upon which are markings, constant in size and arrangement for each species. The great beauty of these markings, together with the infinite variety in the sizes and shapes of the cells of different species, have long made them objects of special study by microscopists. There are said to be upward of ten thousand species.

Anatomy. — A diatom cell is constructed like a box. There is a top and a bottom, known as the upper and lower valve, on both of which markings are found. The valves are connected by membranes known as girdles, or, when detached, as hoops. There are two of these membranes, one attached to each valve, and they are so arranged that one slides over the other just as the rim of a box cover fits over the sides. This arrangement may be seen in Plate I, Figs. *A*, *B* and *C*, where a typical diatom, *Navicula viridis*, is shown in three views. *A* represents the valve* view of the diatom, that is, the view seen when looking directly at the valve or top of the box. *B* represents the girdle* view, the view seen when looking at the girdle or side of the box. *C* is a cross-section through the diatom.

<i>A</i>	<i>B</i>
Valve view.	Girdle view.
Side view.	Front view.
Top view.	Zonal view.
Primary side.	Secondary side.
Secondary side.	Primary side.
Face valvaire.	Face connective.
Vue de profil.	Vue de face.

In consulting books on diatoms the reader should be careful to note the way in which the two views are designated by the author.

* The terms used by different writers to express these two views of a diatom are very confusing. In the following list the terms under *A* represent the valve view and those under *B* the girdle view.

The upper or outer valve is indicated by *a*, and its girdle by *c*. The girdle view shows how this connective membrane of the larger valve fits over a similar one, *c'*, attached to the lower or smaller valve, *b*. These girdles have the power of sliding one upon the other so that the thickness of the diatom, i.e., the distance between the valves, is variable.

The valves of the diatom shown in the figure are covered with furrows or markings, *g*. At the center and at each end there are slight thickenings of the cell wall, known as nodules. The central one is called the central nodule, *d*, and those at the ends, terminal nodules, *e*, *e*. Between these nodules and extending along the medial line of the valve there is a sort of ridge, *f*, in which there is a furrow called a raphé. Through this the protoplasm of the diatom probably communicates with the outer world. The slit is supposed to be somewhat enlarged at the nodules. The raphé, the nodules, and the markings, taken in connection with the shape and size of the valves, are the most important external features of a diatom and are the first to be considered in studying them.

Shape and Size. — There is probably no class of unicellular organisms in which the outlines vary more than in those of the diatoms. From the straight line to the circle almost all the geometrical figures may be found. Some of these may be described as circular, oval, oblong, elliptical, saddle-shaped, boat-shaped, triangular, undulate, sigmoid, linear, etc. The variations in shape are most marked in the valve view. The girdle view, as a rule, is more or less rectangular. The valves are usually plane surfaces with only slight curvatures or undulations. Occasionally the surface is warped as in *Amphiprora* and *Surirella*. As a rule the two valves of a cell are nearly parallel, but in such forms as *Meridion*, *Gomphonema*, etc., the cell is wedge-shaped when seen in girdle view. The most varied forms are found in salt or brackish water, and the common fresh-water forms are so simple and so characteristic that the reader will have little difficulty in assigning them their proper generic names. Some genera have the cell divided more or less completely by internal plates, called septa when fully developed, as in *Rhabdonema*, and designated vittæ when incomplete, as in *Grammatophora*. Some diatoms have external expansions on the margin of the valves, known as alæ or wings, as in *Surirella*; when imperfectly developed, as in *Nitzschia*, they are called keels, and the organism is said to be carinate. These wings or keels usually extend along the border of the raphé. Certain filamentous forms, such as *Melosira*, have processes at the point of attachment. In others these processes are elongated into horns or bristles.

Diatoms vary in size from the minute *Cyclotella*, less than 10 μ in

diameter, to such large forms as *Surirella* and *Navicula*, that sometimes are 1 mm. long. Some filamentous forms grow to a considerable length.

Markings. — The valves of most diatoms are marked with lines or points. In many cases the lines may be resolved into a series of points, pearls, beads or striæ when a higher power of the microscope is used. The variations in the number and size of these points and their uniformity in different individuals of the same species make them convenient objects for testing the resolving power of microscopes.

Cell-contents. — Just inside the cell wall there is a thin protoplasmic lining. This protoplasm sends radiating streams through the cell, and it is possible that a portion of it extends through the openings in the cell wall. It is this layer of protoplasm, also, that secretes the silica of the cell wall. Between the streams of protoplasm (Pl. I, Fig. C) there are what appear to be empty cavities. In or on the borders of these, oil globules may sometimes be observed. There is a nucleus and one or more nucleoli, located near the center of the cell. The most conspicuous portion of the cell contents consists of brownish plates which are usually constant in appearance and position for any particular species. In certain species other internal features have been noted; namely, the contractile zonal membrane, the germinative dot, double nucleus, etc., but of these, little is known.

Physiology. — *External Secretions.* — Living diatoms are covered with a transparent gelatinous sheath, which is probably a secretion from the protoplasm. In many species it is very thin and can be discerned only by the use of stains. In the filamentous and chain-forming species it serves to hold the cells together. In *Tabellaria*, for example, little lumps of the gelatinous substance may be seen at the corners of the cells at the point of attachment. Some species secrete great quantities of gelatinous material and are entirely embedded in it. In a few cases it is of a firmer consistency and forms tubes, stalks, or stipes, upon the ends of which the cells are seated. These stalks attach themselves to stones, wood, etc., immersed in the water.

Movement. — Some of the diatoms exhibit the phenomenon of spontaneous movement. The most peculiar movement is that of *Bacillaria paradoxa*, whose cells slide over each other in a longitudinal direction until they are all but detached, and then stop, reverse their motion, and slide backward in the opposite direction until they are again all but detached. This alternate motion is repeated at quite regular intervals. Some of the free species show the greatest movement, and of these *Navicula* is one of the most interesting. Its motion has been described as a sudden advance in a straight line, a little hesitation, then other rectilinear movements, and, after a short pause, a return upon nearly

the same path by similar movements. The movement appears to be a mechanical one. The diatoms do not turn aside to avoid obstacles, although their direction is sometimes changed by them. The rapidity of their motion has been calculated to be 400 times their own length in three minutes. Their motion shows the expenditure of considerable force. Objects fifty or a hundred times their size are sometimes pushed aside.

Multiplication. — Diatoms multiply by a process of halving or splitting, the Greek word for which gives rise to the name diatom. The cell division is similar to that of all plants, but in this case the process is of especial interest because of the rigid character of the cell walls.

The process begins by a division of the nucleus and nucleolus. The protoplasm expands, forcing the valves apart, the hoops sliding one out of the other. The two halves of the nucleus separate, the pigment collects at either side and a membrane forms, dividing the cell into two parts. Finally the two parts separate. The newly formed membrane becomes charged with silica making a new valve and soon after, its hoop develops. This process is well illustrated by a drawing of J. Deby, shown on Pl. I, Figs. *D*, *E* and *F*. Sometimes the cells separate entirely; sometimes they remain attached, forming filaments, as in *Melosira*, bands as in *Fragilaria*, or zigzag chains as in *Tabellaria*.

The continued process of multiplication results in a constant diminution of the size of the cells. After a certain minimum limit of size has been reached, or after their power of vegetative multiplication has become exhausted, a reproductive process takes place. Usually this consists of a conjugation which results in the formation of large-size auxospores which by multiplication give rise to a new series of cells like the first. In the case of some diatoms reproduction takes place through the formation of microspores which become fertilized by conjugation, and, after a period of rest attain a condition for living an independent life and reproducing in every respect the adult type of mother cell.

KEY TO FRESH-WATER GENERA

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| 1. Valves without a dividing line or cleft; markings more or less radiate; transverse section of cell circular, polygonal, or elliptical, sometimes irregular. | 2 |
| 1. Valves sygomorphous, structure pinnate, not concentric, divided either by a true raphé or by a linear space or line imitating a raphé. | 6 |
| 2. Cells discoid; valves without horns or elevations. | 3 |
| 2. Cells box-like, i.e., with the longitudinal axis greater than in the above; valves with 2 or more elevations or horns. | <i>Eunotogramma</i> |
| 3. Cells short, in long chains. | <i>Melosira</i> |
| 3. Cells disciform, solitary, rarely in short chains. | 4 |
| 4. Valve with two concentric divisions of different structure, one a wide border, the other a central surface. | <i>Cyclotella</i> |
| 4. Valve areolate or punctate with a narrow border of the same structure. | 5 |
| 5. Border with long spines. | <i>Stephanodiscus</i> |
| 5. Border without spines, sometimes echinulate. | <i>Coscinodiscus</i> |

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|--|---------------|
| 6. Valve without a raphé; usually with a pseudo-raphé or median line. | 7 |
| 6. Either one or both valves with a true raphé. | 17 |
| 6. Valves in which the raphé is concealed near the margin on one or both sides of each valve in a more or less elevated keel or wing. | 26 |
| 7. Valve asymmetrical with respect to both the longitudinal and transverse axes; septate not cuneate, finely striated. | 8 |
| 7. Valve symmetrical with respect to the longitudinal axis, asymmetrical to transverse, cuneate, finely striated. | Meridion |
| 7. Valve of varied shape, not cuneate; costate or with transverse rows of puncta. | 9 |
| 8. Cells with two to six nearly straight septa; transverse striae subtly punctate. | Tabellaria |
| 8. Cells not septate but with numerous annuli. | Attheya |
| 9. Valve circular, elliptical to linear, quadrate or cruciform, with transverse costæ; without raphé, a pseudo-raphé sometimes wanting, filamentous. | Diatoma |
| 9. Valve elongate, with small central and terminal elevations, without costæ, but with transverse punctate striae; without genuine central nodule. | 10 |
| 9. Valve lunate; a raphé sometimes partially formed with terminal nodules near the edges. | 16 |
| 10. Cells symmetric in all three directions. | 11 |
| 10. Cells symmetric in only two directions. | 15 |
| 11. Cells with transverse costæ. | Diatoma |
| 11. Cells without costæ. | 12 |
| 12. Striae radiate. | Raphoneis |
| 12. Striae transverse. | 13 |
| 13. Pseudo-raphé broad, lanceolate. | Dimerogramma |
| 13. Pseudo-raphé narrower. | 14 |
| 14. Cells in chains. | Fragilaria |
| 14. Cells solitary or in fascicles. | Synedra |
| 15. Valves asymmetric on transverse axis, ends unequal, cells usually in star-shaped colonies. | Asterionella |
| 15. Valves asymmetric on longitudinal axis, curved. | 16 |
| 16. Cells either free, in fasciae or epiphytic; valve arcuate. | Eunotia |
| 16. Cells solitary or in small clusters, cuneate; valve inflated at one end. | Actinella |
| 17. Valves dissimilar. | 18 |
| 17. Valves similar. | 20 |
| 18. Symmetrical. | Cocconeis |
| 18. Asymmetrical. | 19 |
| 19. In zone view. | Achnanthes |
| 19. To longitudinal axis. | Anorthoneis |
| 19. To transverse axis. | Rhoicosphenia |
| 20. Valves asymmetrical to longitudinal axis. | 21 |
| 20. Valves asymmetrical to transverse axis. | Gomphonema |
| 20. Valves symmetrical. | 22 |
| 21. Valves parallel. | Cymbella |
| 21. Valves not parallel. | Amphora |
| 21. Valves keeled and twisted. | Amphiprora |
| 22. Valves sigmoid. | 23 |
| 22. Valves not sigmoid. | 24 |
| 23. Striae oblique. | Pleurosigma |
| 23. Striae at right angles. | Gyrosigma |
| 24. Striae punctate, nodules elongated. | Frustulia |
| 24. Striae subtly punctate, central nodule forked. | Amphipleura |
| 24. Striae interrupted by blank lines. | Anomoeoneis |
| 24. Striae crossed by longitudinal lines. | Caloneis |
| 24. Striae oblique, median fissures in opposite directions. | Neidium |
| 24. Striae punctate and costate, median line with horns. | Diploneis |
| 24. Striae punctate, central area dilated into a stauros. | Staurois |
| 24. Striae punctate, central area without stauros or horns. | Navicula |
| 24. Striae costate, not punctate. | 25 |
| 25. Valves arcuate. | Epithemia |
| 25. Valves straight. | Pinnularia |
| 26. Valve usually symmetrical, a keel on each border. | Suriella |
| 26. Valve asymmetrical. | 27 |
| 27. Keels of the two valves opposite each other. | Hantzschia |
| 27. Keels not opposite each other. | Nitzschia |

CLASSIFICATION AND DESCRIPTION

This class in most modern classification is considered to have a single family with characters of the class. The family is divided into subfamilies on the presence or absence of surface markings on the valves. The subfamilies are usually arranged in two series on the basis of shape, the first series based on valves approximately circular in outline without a raphé or pseudo-raphé; the second series based on valves showing much longer in one diameter, usually showing a raphé or pseudo-raphé.

SUBFAMILY I. DISCOIDEÆ

Never possessing a raphé or a pseudo-raphé. Cells generally circular or angular, often provided with teeth, spines, or processes. *Stephanodiscus niagara* is the typical form.

TRIBE I. MELOSIREÆ. — Cells cylindrical, adhering and forming a stout filament; valves circular, sometimes armed with spines.

Melosira. — Cells with circular valves and very wide connective bands, attached valve to valve so as to form long cylindrical filaments. In girdle view they are usually rectangular, though sometimes with rounded ends; at the center there are often conspicuous constrictions. The girdles are often marked with dots. The valves are radially striated, with a clear central space. At the edge there is often a keel or row of projecting points, seen in girdle view. There are several common species. *M. granulata* is the most common free-floating form, and *M. varians* the most common filamentous form. (Pl. III, Figs. 15 to 17.)

TRIBE II. COSCINODISCEÆ. — Valves circular, generally with radiating cellules, granules, or puncta; sometimes with marginal or intramarginal spines or distinct ribs; without distinct processes.

Cyclotella. — Cells discoidal, single, occasionally attached valve to valve, but never forming long filaments. Valves circular, finely marked by radial striæ. There is usually an outer ring of radial lines, inside of which there are puncta and fine dots somewhat irregularly arranged. These cannot be seen with low powers. In girdle view the cells appear rectangular or somewhat sigmoidal, with warped valves, as in *C. operculata*. They are often of very small size. (Pl. III, Figs. 18 and 19.)

Stephanodiscus. — Cells discoidal, single. Valves circular, with curved surface, with fringe of minute marginal teeth. Striæ fine radial. Cells rectangular in girdle view, showing projection of middle of valve. Teeth most conspicuous in girdle view. Endochrome conspicuous in rounded lumps. The cells are often of considerable size. (Pl. III, Figs. 20 and 21.)

SUBFAMILY II. FRAGILARIOIDEÆ

Possessing a pseudo-raphé (simple line or blank space) on one or both valves; with or without nodules. Cells generally bacillar, sometimes oval or suborbicular, without processes, spines, or awns. *Synedra gailloni* is the typical form.

TRIBE I. TABELLARIÆ. — Cells with internal plates, or imperfect septa, often forming a filament.

Tabellaria. — Cells square or rectangular in girdle view, attached by their corners and forming zigzag chains. In this view they are seen to be marked with longitudinal dividing plates, which extend from the ends not quite to the middle and which terminate in rounded points. The valves are long and thin, and are dilated at the extremities and in the middle. There are fine transverse striæ and an indistinct pseudo-raphé. The endochrome is usually in rounded lumps. There are two very common species — *T. fenestrata* and *T. flocculosa*. (Pl. III, Figs. 6 to 9.)

TRIBE II. MERIDIONÆ. — Valves symmetrical with respect to the longitudinal axis, asymmetrical to the transverse axis, cuneate, finely striated.

Meridion. — Cells attached valve to valve, forming curved bands seen as fans, circles, or spiral bands. The cells are wedge-shaped, which causes the peculiar shape of the bands. Valves also wedge-shaped, with somewhat rounded ends; furnished with transverse ribs, between which are fine striæ. Pseudo-raphé indistinct. There is one principal species — *M. circulare*. (Pl. III, Figs. 4 and 5.)

TRIBE III. FRAGILARIÆ. — Cells adherent, forming a ribbon-like, fan-like, or zigzag filament, or attached by a gelatinous cushion or stipe.

Epithemia. — Cells cymbiform, symmetrical with respect to the minor axis, with a false raphé and no nodules. Valves marked by lines and pearls approximately at right angles to the major axis, but inclined toward the end of the cell on the convex side. The cells in girdle view are seen to be somewhat inflated at the center. There are several species, differing considerably in the shape of the valves. (Pl. I, Figs. 15 and 16.)

Eunotia. — Cells elongated, symmetrical with respect to the minor axis. Occurring singly, free-swimming or attached. Valves arcuate, with the convex side undulated. Transversely striated, with two false terminal nodules and no medial line. The cells are quadrangular in girdle view. There are but few species, the most common being *E. tridentula*. (Pl. I, Fig. 17.)

Diatoma. — Cells attached by their angles forming zigzag chains, or rarely in bands. In girdle view they are quadrangular. Valves elliptical-lanceolate, with transverse ribs, between which are fine striæ. There is a longitudinal pseudo-raphé. There are two common species — *D. vulgare* and *D. tenue*. (Pl. III, Figs. 1 to 3.)

Himantidium. — Sometimes included under Eunotia. The cells differ from Eunotia by remaining attached after division, forming a band as in *Fragilaria*; by having the convex side of the valve entire instead of undulate; and by being somewhat bent in girdle view. (Pl. II, Figs. 1 and 2.)

Asterionella. — Cells long, linear, inflated at the ends. They are united by their extremities into stars or chains, as shown in the girdle view. The typical group is composed of 8 cells symmetrically and radially arranged. Groups of 4, 6 or 7 are common. When rapidly dividing they may assume a spiral arrangement. The valves are very finely striated, with a straight pseudo-raphé. There is one general species, *A. formosa*, characterized by having the basal end of the cells much larger than the free end, and by having on that end a larger surface in contact with the adjoining cells. There are several varieties, advanced by some authors to the rank of species. The most common is *A. formosa*, var. *gracillima*. (Pl. II, Figs. 3 to 7.)

Synedra. — Cells elongated, straight or slightly curved. Valves somewhat dilated at the center and with a medial line or false raphé and occasionally false nodules. They usually have straight and almost, but not quite, parallel sides. They are

finely transversely striated. There are several common species. *S. pulchella* has lanceolate valves, with ends somewhat attenuated. In girdle view they are seen to be attached valve to valve and present the appearance of a long band or a fine-toothed comb. *S. ulna* has a very long rectilinear valve, with conspicuous transverse striæ. There is a false raphé with a narrow clear space. They are often free-floating. *S. lanceolata* has a long thin valve, swollen at the center, but tapering to sharp points at the ends. *S. radians* has straight needle-like valves. They are united at the base like *Asterionella*, but the cells do not lie in the same plane. (Pl. II, Figs. 8 to 11.)

Fragilaria. — Cells attached side by side, forming bands as in the case of *Synedra pulchella*. Valves elongated, straight, with ends lanceolate or slightly rounded. In girdle view the cells are rectangular and are in contact with each other through their entire length. Valves transversely striated, with a false raphé scarcely visible. There are several common species. (Pl. II, Figs. 12 and 13.)

SUBFAMILY III. NAVICULOIDEÆ

Always possessing a distinct raphé on one or both valves. Central nodule generally present and conspicuous. Cells mostly bacillar in valve view; sometimes broadly oval; without spines or other processes. *Navicula major* is the typical form.

TRIBE I. NAVICULÆ. — Raphé mostly curved. Valves alike, more or less arcuate, cymbiform.

Amphora. — Cells single, ovoidal in girdle view, the girdle often striated or longitudinally punctate. Valves extremely unsymmetrical, with a convex and concave side, with an eccentric raphé, with medial and terminal nodules. The raphé is sometimes near the convex side, sometimes near the concave side, and the medial nodule is often away from the center. There are transverse striæ, radiating somewhat from the medial nodule. This genus is very ornate. There are a number of species, none of them very common in fresh water. (Pl. I, Figs. 1 and 2.)

Cymbella. — Cells generally single, elongated, symmetrical with respect to the minor axis. Valves more or less arched, with one side very convex and the other side slightly or not at all convex; asymmetrically divided by a curved raphé; possessing terminal and medial nodules; marked by transverse bead-like striæ, which do not extend to the raphé, but have a clear space, wider at the medial nodule than elsewhere. There are a number of common species. (Pl. I, Figs. 3 and 4.)

Encyonema. — Cells when young, enclosed in a hyaline mucilaginous tube, in which they multiply by division, pushing each other forward in an alternately inverse position. Valves symmetrical with respect to the minor axis, convex on one side, straight on the other, with rounded extremities that project beyond the straight side. A straight raphé divides the valves into two unequal parts. There are medial and terminal nodules. The striæ are transverse or radiating somewhat from the medial nodule. There is a clear space around the medial nodule, but elsewhere the striæ approach closely to the raphé. There are several species. (Pl. I, Fig. 5.) This group is often considered a subgenus of *Cymbella*.

Cocconema. — Cells when young, borne singly or in pairs on filamentous pedicels which may be simple or branched. They form mucilaginous layers on submerged objects. Later they become free-swimming. The valves are long, large, strongly arched, convex on one side, concave on the other side save for a little inflation in the

middle. The raphé is curved. There are medial and terminal nodules. The striae are rather large pearls, transverse, with very slight radiation, and not approaching the raphé closely. (Pl. I, Fig. 6.) This group is often considered a subgenus of *Cymbella*.

Navicula. — Cells single, symmetrical with respect to both axes. Valves naviculoid, or boat-shaped; of various proportions, some very long and narrow, others short and wide, others ellipsoidal; with straight or slightly curving sides; with ends pointed or rounded. There is a straight raphé with conspicuous medial and terminal nodules. The valves are marked with transverse furrows, that have a slight radial tendency. The cells are rectangular in girdle view and show the nodules plainly. There is a vast number of species and varieties, many of which are very common. (Pl. I, Figs. 7 and 8.)

Stauroneis. — Cells similar to those of *Navicula*. Valves symmetrical, possessing a straight raphé, with medial and terminal nodules. The striae are pearled. There is a narrow clear space along the raphé and a wider transverse clear space at the medial nodule extending to the sides of the valve, so that the valves have the appearance of being marked with a cross. A number of species have been described, but in some instances they are very similar to *Navicula*. (Pl. I, Figs. 9 and 10.)

Frustulia. — Cells quite similar to those of *Navicula*, and enclosed in mucilaginous tubes, as *Encyonema*. Raphé straight, sometimes showing a double line. Striae generally parallel, reaching to the raphé, but not to the central nodule, around which there is a clear space. More common in salt water than in fresh water.

Pleurosigma. — Cells like those of *Navicula*, but with axis turned like a letter S. Raphé sigmoidal. Striae ornate, pearled, very fine on some species. Endochrome in two layers. (Pl. I, Fig. 11.)

Gomphonema. — Cells borne on pedicels more or less branched. Valves wedge-shaped, with more or less undulating margins and rounded ends. A central nodule near the large end. Raphé straight, dividing the valve symmetrically. Striae pearled, transverse, radiating slightly about the nodules. The cells seen in girdle view are wedge-shaped, with straight sides and with central nodule visible. There are a number of species, some of which are common. (Pl. I, Fig. 12.)

TRIBE II. ACHNANTHÆ. — Cells with valves unlike. Valves broadly oval.

Cocconeis. — Cells somewhat arched or lens-shaped; in valve-view, elliptical or discoidal. Striae have a general direction transverse to the axis, but the convexity of the cells gives them the appearance of inclining toward the poles. Upper and lower valves dissimilar, possessing a medial nodule and raphé or pseudo-raphé. (Pl. I, Figs. 13 and 14.)

SUBFAMILY IV. SURIRELLOIDÆ

Always possessing a raphé which is concealed near the margin on one or both sides of each valve in a more or less elevated keel or wing.

Nitzschia. — Cells free, single, elongated, linear, slightly arched, or sigmoidal; with a longitudinal keel and one or more rows of longitudinal points. Valves finely striated, without nodules. There are many species. (Pl. III, Figs. 10 to 12.)

Surirella. — Cells free, single, furnished with alae on each side. A transverse section of the cell shows a double-concave outline. Valves oval or elliptical, with conspicuous transverse tubular striae, or canaliculi, between which there are sometimes very fine pearled striae. There is a wide clear space, or pseudo-raphé. The cells

are sometimes cuneate in girdle view. The valves sometimes have a warped surface. There are many common species, most of them of very large size. (Pl. III, Figs. 13 and 14.)

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CHAPTER XXII

RHODOPHYCEÆ

The Rhodophyceæ are mostly marine except for a few families of primitive or reduced forms which inhabit brooks or portions of rivers with swift currents and rocky bottoms. Apparently they need a relatively large amount of oxygen for their development. The color of the fresh-water forms is variable, the typical red pigment of the group is often poorly developed or masked by other pigments. The filaments are relatively coarse when compared with other fresh-water algæ, and consist of a bundle of small filaments.

Life Cycle. — The male element in sexual reproduction is a small non-motile cell borne on the tip of a highly specialized branch called the antheridium. The corresponding female branch is called the carpogenic branch and consists of a short branch with a small, definite number of cells, bearing the egg cell at the tip. The egg cell is provided with a long protuberance or trichogyne, on its distal end, which reaches to the surface of the plant. The spermatia or non-motile male cells are washed against the trichogyne, where the male nucleus enters the egg cell and fertilizes the female nucleus. The fertilized egg then buds off whorls of reproductive cells called carpospores which are liberated. These germinate and reproduce the vegetative portion of the plant.

KEY TO FRESH-WATER GENERA

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|---|------------------------|
| 1. Main axis distinctly articulate, except in oldest parts. | 2 |
| 1. Body of frond not distinctly articulate. | 3 |
| 2. Main axis of a single series of large cells, later developing a cortex of 1 to 4 layers of minute cells; slender, much branched; not gelatinous. | <i>Compsopogon</i> |
| 2. Main axis of a single series of cells, at first naked, later with rhizoidal coating; gelatinous. | <i>Batrachospermum</i> |
| 3. Gelatinous; axis of very slender, interwoven filaments, surrounded by a dense coating of long, articulate hairs. | <i>Thorea</i> |
| 3. Not gelatinous; no hairs. | 4 |
| 4. Elongate, tubular, slender, more or less branched, with firm, cellular wall and slender central axis; usually with regular, torulose swellings. | <i>Lemanea</i> |
| 4. Short, nearly solid, densely branched, with irregular swellings. | <i>Tuomeya</i> |

Batrachospermum (Pl. XIX, Fig. 7) and *Lemanea* are the genera most frequently encountered. The others are less well known and mostly tropical.

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CHAPTER XXIII

FUNGI

Fungi are flowerless plants in which the special characteristic is the absence of chlorophyll and starch. Lacking these, they are unable to assimilate inorganic matter, and consequently live a saprophytic or parasitic existence, that is, they live upon dead organic matter or in or upon some living host. They are essentially terrestrial plants but some of them live a sort of semi-aquatic life.

Many very different forms are included among the fungi. On the one hand there are microscopic forms; among these most authors include the bacteria, because they have no chlorophyll; on the other hand there are the mushrooms which are often very large. Fungi usually consist of two parts, the mycelium or vegetative body and the reproductive organs or fruit. The mycelium is a mass of delicate, often septate, branched, usually colorless filaments intertwined to form a cottony or felty layer. It is the spawn of mushrooms and the common mold or mildew seen on decaying vegetable matter. The reproductive bodies are mycelial filaments bearing spores of various kinds in various positions. It is by differences in the method of fruiting that the different fungi are distinguished from each other.

Classification. — Six classes of fungi are usually recognized.

1. **SCHIZOMYCETES** (fission fungi or bacteria). Typically unicellular plants, cells small and relatively primitive in organization. The cells are of many shapes, spherical, cylindrical, spiral or filamentous; cells often united into groups, families or filaments, occasionally in the latter showing some differentiation among the cells, simulating the organization of some of the Myxophyceæ. Multiplication typically by cell fission. Endospores formed by some species.

2. **MYXOMYCETES**. — A group equally well classed among the Rhizopoda on the basis of the vegetative cells, but these cells uniting into a plasmodium and forming spores. Fructifications resembling puff-balls, with which they were first classified. No aquatic species known.

3. **PHYCOMYCETES**. — Mycelium non-septate, with many nuclei present in a single cell. Reproduction by means of motile zoöspores, gametes or conjugation.

4. **ASCOMYCETES**. — Mycelium septate, cells usually uni- or bi-nucleate. Reproduction by means of spores discharged from a specialized cell known as an ascus. Practically none aquatic.

5. **BASIDIOMYCETES**. — Mycelium septate, cells usually uni- or bi-nucleate. Reproduction by means of spores borne on a specialized cell known as a basidium. Practically none aquatic.

6. **FUNGI IMPERFECTI.** — This large and heterogeneous group includes all the fungi whose complete life cycle is unknown. Reproduction various, mostly by conidia, no ascospores, basidiospores or sporangia known.

With exception of the iron bacteria the fungi as a group are seldom seen in clean water. They are more common in sewage, but the number of important genera occurring is small. Apart from the Schizomycetes and Phycomycetes discussed in the two succeeding chapters the following organisms are at times encountered by sanitary biologists: The molds *Aspergillus* (Pl. X, Fig. 7), *Penicillium* (Pl. X, Figs. 5 and 6), and *Mucor* (Pl. X, Fig. 8) and the yeast *Saccharomyces* (Pl. X, Fig. 4).

CHAPTER XXIV

SCHIZOMYCETES

The Schizomycetes or bacteria are mostly unicellular plants, with small cells, rather primitive in organization. The cells vary much in shape, multiplication is typically by fission although in some groups resting spores are produced under conditions unfavorable for vegetative growth. The more highly developed forms are filamentous, at least in favorable surroundings. The strictly unicellular forms are very numerous and are not discussed in the present work. The higher bacteria are divided into four orders only two of which are important to the water analyst.

CHLAMYDOBACTERIALES

The Chlamydobacteriales or Iron Bacteria are distinguished from the other filamentous orders by the presence of a thick gelatinous sheath in which iron is usually deposited.

KEY TO GENERA

- | | |
|--|---------------------|
| 1. Filaments usually not permanently attached. | 2 |
| 1. Filaments attached. | 3 |
| 2. Filaments straight or at least not twisted. | <i>Leptothrix</i> |
| 2. Filaments twisted. | <i>Didymohelix</i> |
| 3. Filaments unbranched. | <i>Crenothrix</i> |
| 3. Filaments showing pseudo-dichotomous branching. | 4 |
| 4. Swarm cells developed. Usually without iron in sheath. | <i>Sphaerotilus</i> |
| 4. Spherical, non-motile conidia, usually with iron in sheath. | <i>Clonothrix</i> |

Leptothrix. — Filaments of cylindrical, colorless cells, with a sheath at first thin and colorless, later thicker, yellow, or brown, becoming encrusted with iron oxide. The iron may be dissolved by dilute acid, whereupon the inner cells show up well. Multiplication is through the division and abstraction of cells, and motile cylindrical swarm cells. Swarm cells sometimes germinate in the sheath, giving the appearance of branching. Pseudo-dichotomous branching may occur. *Leptothrix ochracea* (Leiblein) Kützing is the best-known species. (Pl. IV, Fig. 1.)

Crenothrix. — Filaments unbranched, showing differentiation of base and tip, attached, usually thicker at the tip. Sheaths plainly visible, usually colorless, becoming brownish from iron oxide in old filaments. Cells cylindrical or spherical. Multiplication by non-motile spherical gonidia; cells dividing in three planes to form gonidia. *Crenothrix polyspora* Cohn is the only well-known species. (Pl. IV, Fig. 4.)

Didymohelix. — Filaments twisted, or two filaments twisted together. Young cells colorless, later brown to rust-red through deposition of iron. Simple filaments

show no division into cells even when iron is removed with acid and stain applied. Sheath not demonstrable. *Didymohelix ferruginea* is the type species; also known as *Gallionella ferruginea*, *Chlamydothrix ferruginea*.

Sphaerotilus. — Attached colorless threads showing false branching, making a pseudo-dichotomy. Filaments consist of rod or oval cells, surrounded by a thin, firm sheath. Multiplication occurs both by non-motile and motile gonidia, the

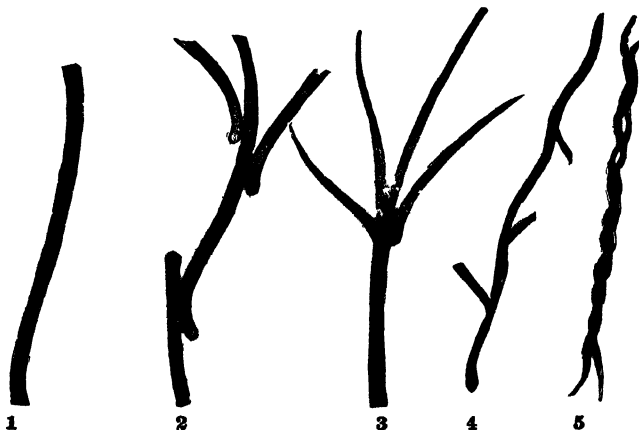


FIG. 123. — Iron Bacteria. 1. *Crenothrix*; 2. *Sphaerotilus dichotomus*; 3. *Clonothrix*; 4 and 5 *Didymohelix*.

latter with a clump of flagella near one end. *S. natans* is the type. (Pl. C, Fig. 4.) *Sphaerotilus dichotomus* (*Cladothrix dichotoma*) is another well-known species, which is differentiated by false branching from *Sphaerotilus natans*, the so-called sewage fungus. (Pl. IV, Fig. 2.)

Clonothrix. — Filaments with false, dichotomous or irregular branching, attached with contrast of base and tip, thicker at the base and tapering to the tip. Sheath always present, thin on young filaments, later becoming encrusted with iron and manganese. Multiplication by small, non-motile conidia of spherical form, formed from the disk-shaped cells near the tip by longitudinal division and rounding up. The type species is *Clonothrix fusca*.

THIOBACTERIALES

The Thiobacteriales are distinguished by the cells either containing granules of free sulphur or bacteriopurpurin or both, usually growing best in the presence of hydrogen sulphide. Spores rarely or never formed

KEY TO FRESH-WATER GENERA

- | | |
|---|--------------------|
| 1. Cells containing bacteriopurpurin with or without sulfur granules. | 2 |
| 1. Cells containing sulfur granules but lacking bacteriopurpurin. | 17 |
| 2. Cells without sulfur granules. | 3 |
| 2. Cells containing sulfur granules. | 7 |
| 3. Cells spherical or short rods. | 4 |
| 3. Cells rod-shaped, many embedded in the same slimy capsule. | Rhodocystis |

3. Cells free and elongate.	5
4. Cells in chains, each chain surrounded by a capsule.	Rhodonostoc
4. Cells free.	Rhodospheera
5. Cells bent or curved.	6
5. Cells not bent, motile.	Rhodobacillus
6. Cells short, comma-shaped, with a single polar flagellum.	Rhodovibrio
6. Cells spiral, with spiral flagella.	Rhodospirillum
7. Cells united, at least during a part of the life history, into families.	8
7. Cells free, capable of swarming at any time.	16
8. Cell division such that masses of cells, not merely plates, are formed.	9
8. Cell division in two planes forming plates of cells.	13
8. Cell division in one plane.	14
9. Cell division in three planes.	10
9. Cell division at first in three planes, then in two.	Lamprocystis
10. Cells capable of swarming.	11
10. Cells not capable of swarming.	12
11. Families small compact, enclosed singly or several together in a cyst.	Thiocystis
11. Cells large, 7 to 8 μ , loosely bound by gelatin into families.	Thiospheera
12. Spherical cells spread out upon the substratum in flat families loosely enveloped in a common gelatin.	Thiocapsa
13. Arranged in regular packets like Sarcina.	Thiosarcina
13. Cells occurring regularly in fours.	Lampropedia
13. Cells occurring in a film or membrane and regularly disposed in tetrads.	Thioderma
14. Cells connected by plasma threads, families amœboid, motile.	Amœobacter
14. Cells arranged in a net united by their ends.	Thiodictyon
14. Cells arranged otherwise.	15
15. Capable of swarming, cells loosely aggregated in gelatin.	Thiothece
15. Non-motile, cells appressed in a colony.	Thioplococcus
16. Cells spiral.	Thiospirillum
16. Cells spindle-shaped.	Rhabdomonas
16. Cells cylindrical.	Chromatium
17. Filamentous forms.	18
17. Unicellular, motile forms.	19
18. Filaments non-motile, with a contrast between base and tip, attached.	Thiothrix
18. Filaments motile (oscillating) not attached, not differentiated into base and tip.	Beggiatoa
19. Cells longer, very large, 42 to 86 μ , with peritrichous flagella.	Hillhousia
19. Cells ellipsoidal (spherical when newly divided), containing granules of calcium oxalate or perhaps sulfur.	Achromatium

Beggiatoa. — Filaments colorless, containing numerous dark sulphur granules, short, 1 to 3 μ in diameter, exhibiting an oscillating movement similar to that of *Oscillatoria*. The most common species is *Beggiatoa alba* (Vaucher) Trevisan. (Pl. IV, Fig. 3.)

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CHAPTER XXV

PHYCOMYCETES

The Phycomycetes as the name implies show many close relationships to certain groups of algæ, especially the Chlorophyceæ. The mycelium is cœnocytic, septation occurring only in cutting off the reproductive cells or when the plant is growing in very unfavorable environment. The cells are still cœnocytic even when the mycelium is septate.

Reproduction. — The phycomycetes reproduce by both non-sexual and sexual means.

Non-sexual Reproduction. — On the approach of conditions unfavorable for growth, a thick wall is formed around a portion of a hypha and a spore is formed, known as a chlamydospore, or, if more highly specialized, as a gemma. Special organs known as sporangia are often produced which contain spores. These spores are formed by progressive cleavage of the protoplasm until all of it is used up. They may be motile as in the Saprolegniales or non-motile as in the Mucorales. The motile sporangiospores, or zoöspores, usually have one or two flagella, a blepharoplast at the base of the flagellum, a pigment spot which is sensitive to light rays, and a densely-staining nucleus. These spores are capable of settling down, germinating, and reproducing the plant.

Sexual Reproduction. — In the first series one may find all gradations between conjugation of equal flagellated gametes to highly differentiated sperms and eggs. In the second series, two filaments of mycelium come together, fuse and produce a sexual resting spore, which later germinates to form the vegetative filaments similar to the conjugation in Spirogyra.

CLASSIFICATION AND DESCRIPTION

The group is usually divided into two series having little relation to each other. The first series is characterized by having motile zoöspores or gametes, the second series by having conjugation of the filaments as in Spirogyra among the algæ.

CHYTRIDIALES. — This order consists of very simple forms which are considered as either primitive or degenerate, found mostly as minute parasites on algæ or other plants.

MONOBLEPHARIDALES. — Plant slender, branched, without a distinct stalk; not constricted into joints; egg single, usually sculptured, in some species ripening out-

side the oögonium, fertilized by an active sperm with one flagellum. Spores uniflagellate, monoplanetic.

BLASTOCLADIALES. — Plant with a distinct enlarged stalk or, if the base is not distinctly differentiated, then branched in a dichotomous or verticillate way; peculiar resting cells present which are probably parthenogenetic eggs. These have thick, brown, distinctly pitted walls and completely or almost completely fill the thin-walled oögonium, out of which they often slip at maturity. Antheridia unknown. Spores with one or two flagella monoplanetic.

LEPTOMITALES. — Plant often with a distinct stalk, the mycelium constricted at intervals into joints, which in the vegetative region are usually connected by small channels through the nodes; oögonia and antheridia present in most species, the egg always single and with periplasm, often with a sculptured surface; spores monoplanetic.

KEY TO GENERA OF LEPTOMITALES

1. Main axis somewhat larger but not otherwise differentiated from the branches, usually segmented
Sexual organs not present. 2
1. Main axis differentiated from the branches, usually not segmented. Sexual organs present. 4
2. Segments short, sporangia mostly elongate, broadest at the base, renewed by proliferation within the old sporangium; zoöspores uniflagellate. **Gonapodya**
2. Segments cylindric, sporangia not renewed by proliferation; zoöspores biflagellate. 3
3. Sporangia not differing from the segments in form, zoöspores swarming immediately; resting spores absent. **Leptomitius**
3. Sporangia pyriform or ellipsoidal, broader than the segments, zoöspores collected at the mouth of the sporangium before swarming. **Apodachlya**
4. Rhizoids weakly developed or absent. **Sapromyces**
4. Rhizoids abundant. 5
5. Sporangia elongate-ellipsoidal, often covered with spines. **Araispora**
5. Sporangia broad-ellipsoidal, always smooth. **Rhiphidium**

Leptomitius. — Hyphæ delicate, sparingly branched apically and soon appearing dichotomous, constricted at intervals into distinct segments with a conspicuous cellulose plug separating them. Sporangia apical and then in rows in basipetal succession. Spores in a single row. This genus is often found in masses in pipes conveying sewage or on the banks of polluted streams. (Pl. XI, Fig. 3.)

SAPROLEGNIALES. — Mycelium not constricted into joints at intervals; oögonia containing one or, more often, several eggs in the formation of which all the protoplasm of the oögonium is used; eggs always smooth, not completely filling the oögonium except in *Leptolegnia*; antheridia present in most species, but even when present fertilization is not always effected; asexual spores biflagellate, dipplanetic or monoplanetic.

KEY TO GENERA OF SAPROLEGNIALES

1. Sporangia rare or absent; oögonia with very thick pitted walls, the antheridia arising from immediately below them and running up their sides. **Aplanes**
1. Not as above. 2
2. Spores normally leaving the sporangium by a common mouth. 3
2. Spores not leaving the sporangium by a common mouth. 8
3. Spores all (normally) swarming separately on escaping from the sporangium. 4
3. Spores all collecting in a hollow sphere or an irregular group at the mouth of the sporangium on escaping. 7
3. Part of the spores on emerging swimming away or sluggishly jerking away and encysting separately from the remainder, which stop at the sporangium mouth. Sporangia rounded at the tip and not tapering, in great part proliferating cymosely as in *Achlya* but at times filaments may grow through empty sporangia as in *Saprolegnia*. Eggs centric. **Protoachlya**

- | | |
|---|----------------------|
| 4. Sporangia not thicker than the vegetative hyphæ; zoöspores in a single row. | Leptolegnia |
| 4. Sporangia thicker than the hyphæ; zoöspores not in a single row. | 5 |
| 5. New sporangia formed within the empty ones. | Saprolegnia |
| 5. New sporangia formed in greater part by cymose branching. | 6 |
| 6. Antheridia on every oogonium, androgynous. | Pythiopsis |
| 6. Antheridia absent or on less than half the oogonia, dichinous. | Isoachlya |
| 7. Sporangia usually thicker than the vegetative hyphæ; zoöspores not in a single row. | Achlya |
| 7. Sporangia thicker than the vegetative hyphæ; zoöspores in a single row. | Aphanomyces |
| 8. Spores encysting within the sporangium, then emerging separately through the sporangium wall and swarming. | Dictyuchus |
| 8. Spores set free by the breaking up of the sporangium wall. | Thraustotheca |

Saprolegnia. — Saprophytic or parasitic on plants or animals in water, sometimes producing pathologic conditions as for example in the "salmon-disease." They are often seen on dead flies, etc. The zoöspores are biflagellate, and motile even within the sporangium. As soon as they are discharged they swim away, soon coming to rest and encysting in spherical form; after a few hours they emerge again through a minute opening in the cyst and swim away more actively, finally coming to rest on a nutrient substratum, if available, and sending into it a slender tube which grows and branches into the mycelium. (Pl. XI, Fig. 1.)

Achlya. — Closely resembling *Saprolegnia* in size, growth, and appearance of vegetative parts. Zoöspores on leaving the sporangium coming to rest in a hollow sphere or irregular cluster, encysting there and after a few hours swimming again as in *Saprolegnia*. (Pl. XI, Fig. 2.)

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CHAPTER XXVI

PROTOZOA

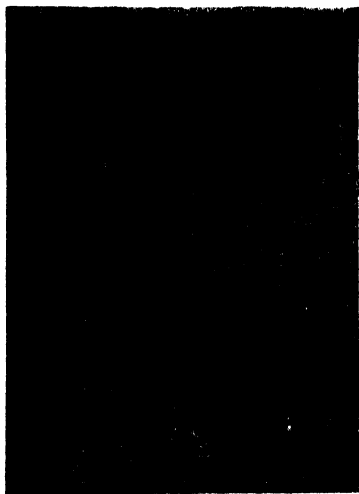
The Protozoa are the lowest organisms belonging to the animal kingdom. The name protozoa was used by the early writers to describe all minute organisms, whether animal or vegetable, but of late it has come to have a more definite meaning. It is now applied to those animal forms which are unicellular or multicellular by aggregation. Structurally the protozoa are single cells, and where there is an aggregation of several cells each one preserves its identity. There is no differentiation, no difference in the function of the different cells. Thus, the protozoa are definitely set off from the metazoa or enterozoa, which are multicellular, and which have two groups of cells, one group forming the lining to a digestive cavity and the other group forming the body wall, which differ both in structure and in function. Most of the protozoa are strictly unicellular.

It is extremely difficult to separate the unicellular protozoa from the unicellular protophyta. Theoretically there is a sharp distinction between the animal and vegetable kingdoms. Definitions may be found applicable to the higher types of life, but they overlap and become confused when applied to the lowest forms. For example, the fundamental difference between the two kingdoms is supposed to lie in the phenomenon of nutrition. Plants can take up the carbon, oxygen, hydrogen, and nitrogen from mineral matter dissolved in water — the nitrogen in the form of ammonia or nitrates, the carbon in the form of carbonic acid. Their food is in solution; hence they need no mouth or digestive apparatus. They absorb their nourishment through their entire surface. Animals, however, cannot take up nitrogen in a lower state than is found in the albumens, nor carbon except in combination with oxygen and hydrogen in the form of fat, sugar, starch, etc. The albumens and fats are not soluble in water; consequently the food of animals must consist of more or less solid particles. Animals therefore require a mouth, digestive cavity, organs for obtaining their food, etc. As albumens, fats, etc., are found in nature only as products of plant or animal life, it follows that all animal life is dependent upon vegetable life or other animal life. There are, however, certain plants that live on organic matter (insectivorous plants, pitcher plants) and even have digestive cavities, but all their relations show that they are real plants. There are other plants that are devoid of chlorophyll (fungi), yet no one

would think of calling them animals. Then there are many unicellular organisms that contain chlorophyll and have the vegetable, or holophytic, mode of nutrition, but that resemble the animal kingdom in other respects. Such, for example, are the Dinoflagellata and many of the green Flagellata. Because it is difficult to draw a sharp line between the vegetable and animal unicellular forms Hæckel proposed a new group, the Protista, lying between the two kingdoms. This group has been since known as the Phytozoa. The term is not used in this work but the organisms have been placed in the one or the other of the two kingdoms according to the best available authority.

The Protozoan Cell. — The protozoan cell, or the individual protozoön, is a single mass of sarcode, or protoplasm, that possesses in a general way all the properties of the protoplasm of higher animal cells. It has a certain amount of irritability and movement, it assimilates food, it grows, and reproduces its kind. It is subject to the same chemical and physical reactions that are observed in higher forms. In size it varies from the tiniest corpuscle to a mass an inch in diameter. It is irregular in form, without a definite boundary; or it has a cell wall and a definite symmetrical outline. Internally the cell usually contains a solid nucleus or a nuclear substance distributed through the cell and recognized by staining. In fresh-water forms it usually contains a contractile vacuole, which may be seen to expand and contract, discharging a watery or gaseous matter through the cell. There are also permanent vacuoles of watery fluid, gastric vacuoles formed by the water taken in with the food, oil globules, and solid particles of starch, chlorophyll, etc. Externally there may be a cortical substance — a denser layer of protoplasm giving definite shape to the cell — that is sometimes contractile. The exterior protoplasm may contain such secreted products as chitin, a nitrogenous horny matter, or cellulose, a non-nitrogenous substance, forming a cell wall, cell cuticle, or matrix. Substances may be deposited even outside of the protoplasmic layer. If perforated, they are known as shells; if closed entirely, as cysts. Cysts are usually of a horny nature and are temporary products. External secretions of calcium carbonate, silicates, etc., are sometimes present.

The cell protoplasm often exhibits certain internal flowing movements, described as the "streaming of the protoplasm." Portions of the protoplasm often extend outward, forming processes. These are of two kinds, and the distinction between them has been used as a basis of classification. Those protozoa that have lobose, filamentous processes, known as pseudopodia, are called myxopods; those that have motile hair-like processes, known as cilia or flagella, are called mastigopods.



Cylindrospermum and Vorticella.



Melosira.



Mallomonas, etc.



Acineta.

PLATE G.

Photomicrographs of Microscopic Organisms. Diatomaceæ, Cyanophyceæ
and Protozoa.

The simplest protozoa absorb solid particles of food at any point on their surface. Digestion takes place within the cell. Protozoa higher in the scale of life have a distinct oral aperture through which the food enters, a sort of pharyngeal passage, and an anal aperture through which undigested portions of food are expelled. There is no real digestive cavity. Some protozoa exhibit a simple kind of respiration. Experiment has shown that they take up oxygen and give out carbonic acid. Multiplication takes place by binary division, by unequal division, i.e., budding or gemmation, and by multiple division or spore formation. Strictly there is no sexual reproduction, though in certain instances there are processes corresponding to it.

Various classifications have been suggested for the protozoa. None are entirely satisfactory. Bütschli has divided the protozoa into four classes: the Sarcodina, Sporozoa, Mastigophora and Infusoria.

Calkins has suggested that the four main groups of protozoa should have the taxonomic value of sub-phyla. His recent classification in general is followed below so far as it relates to the forms with which the water analyst is concerned. Many families and some entire orders are omitted.

KEY TO GENERA OF PROTOZOA DESCRIBED

Protozoa

Locomotion by means of pseudopodia.....	<i>Sarcodina</i>
Locomotion by means of flagella.....	<i>Mastigophora</i>
Locomotion by means of cilia.....	<i>Infusoria</i>
Parasitic forms without any of the above structures.....	<i>Sporozoa</i>

Sarcodina

1. Pseudopodia variable in form — not radiating or still.	<i>Rhizopoda</i>
1. Pseudopodia delicate, radiating, stiff.	<i>Actinopoda</i>
2. No "central capsule."	<i>Heliozoa</i>
2. With a "central capsule" (marine only).	<i>Radiolaria</i>
3. Naked, large, several pseudopodia.	<i>Amœba</i>
3. With test or shell.	4
4. Shell of one piece, chitinous, no spines.	<i>Arcella</i>
4. Shell of one piece, variable number of spines.	<i>Centropyxis</i>
4. Shell covered with sand, mud, etc.	<i>Diffugia</i>
4. Test dome-shaped, circular in cross-section, without spine.	<i>Euglypha</i>
4. Test dome-shaped, ellipsoidal in cross-section, aperture circular.	<i>Trinema</i>
5. Without skeleton or outer envelope.	6
5. Outer envelope or skeleton present.	7
6. Vacuolated ectoplasm not sharply separated from endoplasm.	<i>Actinophrys</i>
6. Vacuolated ectoplasm sharply separated from endoplasm.	<i>Actinosphaerium</i>
7. Envelope jelly-like, radial surface markings.	<i>Heterophrys</i>

Mastigophora

1. With cellulose shell and 2 flagella — one directed backwards; the other lying in a transverse groove.
Order *Dinoflagellida* 2
2. Without membrane surrounding body and with cross furrow extending wholly around body.
Gymnodinium

2. With delicate one-piece cellulose shell.	Glenodinium
2. Cuirass with or without horn-like processes.	Peridiniidae 3
3. Horns two or three in number.	Peridinium
3. One anterior, one to three posterior horns.	Ceratium
4. Colonial forms.	5
4. Solitary forms.	10
5. Colony branching or stalked.	6
5. Colony spherical.	7
6. Colony sometimes colored, stalked, no "collar."	Anthophysa
6. Green or greenish-brown, free-swimming.	Dinobryon
7. Individuals reaching center of sphere.	8
7. Individuals peripherally placed and embedded in jelly.	9
8. Without test, colonial, in jelly.	Syncrypta
8. Colonies globular, yellowish or brownish-green, individuals loosely attached.	Synura
9. Colonies of very numerous individuals, spheres inside	Volvox
9. Individuals fixed by inner gelatinous branched processes, no spheres inside.	Uroglenopsis
10. Colored, usually green.	11
10. Colorless.	13
11. Body rigid with thick shell or pellicle	12
11. Body flexible, showing "euglenoid" movements.	Euglena
11. Flexible, flattened with pointed caudal process.	Phacus
12. With siliceous plates and needles.	Mallomonas
12. Anterior end truncated, with furrow; 2 lateral, yellow chromatophores	Cryptomonas
12. With 2 flagella, close-fitting cellulose membrane, spherical to ellipsoidal, one large chromatophore.	Chlamydomonas
	Trachelomonas
	Chilomonas
	Raphidomonas
	Monas
	Cercomonas
12. With brownish shell, often spiny, 1 flagellum.	
13. 2 flagella from center of anterior end.	
13. 2 flagella, one trailing; with trichocyst-like rods.	
13. Very minute, 2 active flagella.	
13. 2 flagella; 1 anterior, 1 posterior.	

Infusoria

1. Only young forms with cilia, adults with peculiar tentacles.	Suctorina 2
1. Ciliated throughout life, no tentacles.	Ciliata 3
2. Pellicle delicate, test without free margin and membrane-like tentacles in fascicles.	Acineta
2. Mouth with zone of membranelles.	5
3. No such zone of membranelles.	Holotrichida 4
4. Mouths usually closed, oral membrane absent.	6
4. Mouths usually open, oral membrane present.	7
5. Entire body ciliated, membranelles on left margin of peristome.	Heterotrichida 8
5. Not stalked, few cilia or none, membranelles are motile organs	Oligotrichida 9
5. Body flattened, cilia and cirri on ventral surface only.	Hypotrichida 10
5. Stalked forms; single, almost complete circle of membranelles turning to right.	Peritrichida 11
6. With sculptured shell.	Coleps
6. Elongated, mouth surrounded by a circle of longer cilia.	Trachelocerca
6. Body flask-shaped, mouth region sharply truncated.	Enchelys
6. Ellipsoidal, mouth surrounded by basketwork of trichites.	Nassula
7. Dorsal side arched, mouth anterior, membrane small.	Colpidium
7. Ellipsoidal, membrane sail-like.	Pleuronema
7. Slipper-shaped, mouth near middle of body.	Paramecium
8. Body trumpet-shaped.	Stentor
8. Body large, frontal field deeply insunk, sac-like.	Bursaria
9. Spheroidal, central girdle of bristle-like cilia.	Halteria
9. With rigid chitinous test open at both ends.	Tintinnus
9. Test with anterior decorations, anterior openings slight or absent.	Codonella
10. With cirri only; lateral, ventral, frontal and anal cirri present.	Euplotes
11. Cells with contractile stalks.	12
11. Cells without contractile stalks.	13
12. Solitary; a single highly contractile stalk.	Vorticella
12. Individual stalks contract separately; not connected.	Carchesium
12. Entire colony contracts; stalk threads connected.	Zoothamnium
13. Peristome disk not stalked; feather-like colonies.	Epistylis

CLASSIFICATION AND DESCRIPTION

SUB-PHYLUM SARCODINA

There are two classes — Actinopoda and Rhizopoda. The Actinopoda include the two subclasses Heliozoa and Radiolaria. The latter consists of marine forms and with the subclass Foraminifera of the Rhizopoda are omitted.

SUBCLASS HELIOZOA

Actinopoda, generally spheroidal in form, with numerous radial, filamentous pseudopodia which ordinarily exhibit little change of form, though they are elastic and contractile. Protoplasm richly vacuolated. One or more nuclei. Symbiotic forms often occur in the endoplasm. The Heliozoa are generally found in fresh water. They are closely related to the marine Radiolaria.

Actinophrys. — A spherical mass of colorless protoplasm seemingly filled with small bubbles, with numerous long, fine rays springing from all parts of the surface. Contractile vesicle large and active. The organism moves with a slow gliding motion. It feeds on smaller protozoa, algæ-spores, etc. The most important species is *A. sol*, otherwise known as the "sun-animalcule." It is very common in swamp water. (Pl. XI, Fig. 10.)

Actinosphærium. — Spherical, relatively large form, 1 mm. or more in diameter. The ectoplasm and endoplasm are sharply differentiated, the ectoplasm being coarsely vacuolated. The axial filaments of the pseudopodia project but slightly into the border of the ectoplasm. Numerous nuclei are present in the endoplasm. There are from 2 to 14 contractile vacuoles in the ectoplasm. Actinosphærium is omnivorous, its food consisting of the larger infusoria or small metazoa such as rotifers. It is very widely distributed in fresh water, particularly where there is a considerable quantity of decaying organic material.

Heterophrys. — Like Actinophrys in general form, but with the body enveloped with a thick stratum of protoplasm defined by a granulated or thickly villous surface and penetrated by the pseudopodal rays.

CLASS RHIZOPODA

Protozoa provided with variable, retractile, root-like processes or pseudopodia. Great majority with tests of pseudo-chitin on which mineral substances are cemented. Majority are multinucleate. Contractile vacuoles present in fresh-water forms.

There are four subclasses — Proteomyxa, Mycetozoa, Foraminifera and Amœbæa. Of these the forms most common in fresh water belong to the Amœbæa.

SUBCLASS AMŒBÆA

Rhizopoda in which the "amœba-phase" predominates in permanence and physiological importance. Pseudopodia lobose, not filamen-

tous, arborescent or reticulate. A denser external layer of protoplasm usually noticed. Provided with one or more nuclei and usually with a contractile vacuole. Reproduction commonly effected by simple fission, sometimes by a kind of budding.

Amœba. — A soft, colorless, granular mass of protoplasm; possessing extensile and contractile power; devoid of investing membrane, but having an external thickening or protoplasm; with variable, lobose, finger-like processes; ingesting food by flowing around and engulfing it; the absorbed food material (diatoms, algæ, etc.) is often conspicuous. There are several species that vary in size and in the character of the pseudopodia. A common habitat is the superficial ooze of ponds or ditches. (Pl. XI, Fig. 4.)

Arcella. — An amœba-like organism enclosed in a chitinoid shell that is variable in shape, but more or less campanulate or dome-shaped, and that has a circular, somewhat concave base. When seen from above, it is disk-shaped, with a pale circular spot in the middle; when seen from the side, the upper surface is strongly convex. The shell usually has a brown color, and is sometimes smooth and sometimes hexagonally marked. The protoplasmic mass occupies the central portion of the shell, but pseudopodia project through an opening in the concave base. There are many species, differing in shape and in the marks, ridges, etc., on the shell. *A. vulgaris* is the most common. (Pl. XI, Figs. 5 and 6.)

Centropyxis. — Shell covered with foreign bodies, cap-shaped, with several spines at the apical pole. Mouth eccentric in position. Pseudopodia finger-form. Length of shell 80 to 260 μ ; height, 36 to 80 μ ; breadth, 72 to 220 μ . Centropyxis is found very commonly in swamp or bog water along with algæ.

Diffugia. — Body enclosed in a spherical or pear-shaped membrane in which sand grains, etc., are embedded. The lower part is sometimes prolonged as a neck, at the end of which is situated the mouth, through which finger-like pseudopodia may project. The surface of the shell is very rough and usually has a brownish or a gray color. Diatoms, etc., are frequently attached to the shell. The contained protoplasmic mass frequently has a green color, but the pseudopodia are colorless. There are several species, varying in shape and size. The diameter of Diffugia shells varies from 35 to 300 μ . (Pl. XI, Fig. 7.)

Euglypha. — Body enclosed in a hyaline, ovoid shell, composed of regular hexagonal plates of chitinoid membrane, arranged in alternating longitudinal series. At the mouth the plates form a serrated margin. The upper portion of the shell is sometimes provided with spines. The protoplasm is almost entirely enclosed by the shell; the pseudopodia are delicate and branched. There are several species. (Pl. XI, Fig. 8.)

Trinema. — Body enclosed in a hyaline, pouch-like shell, with long axis inclined or oblique, and with mouth subterminal. Dome rounded; mouth inverted, circular, beaded at border. Pseudopodia as in Euglypha, but fewer in number. The two genera are quite similar, but Trinema is usually much smaller. One species. (Pl. XI, Fig. 9.)

SUB-PHYLUM MASTIGOPHORA

Protozoa bearing one or more lash-like flagella, occasionally supplemented by cilia, pseudopodia, etc. With an indistinct, diffuse, or definite ingestive system, and usually with one or more contractile

vesicles. Multiplication takes place by fission and by sporulation of the entire body mass, the process often being preceded by conjugation of two or more zooids. There are two classes — Phytomastigoda and Zoömastigoda.

CLASS I. PHYTOMASTIGODA

This class includes the flagellated forms that contain chlorophyll and also those that, though colorless, show by their structure and life history a close relationship with the chlorophyll-bearing organisms. The great majority of these forms are from 25 to 100 μ in length although some are extremely small. Many of the forms are amœboid while some, as the Euglenida, undergo changes in form without breaking the periphery. Such organisms are said to be metabolic.

Flagella vary in number from 1 to 4. In many cases one (primary) flagellum is directed forward while a second one, known as a trailing flagellum, is directed backward. Nuclei are chiefly simple vesicular and endosome-bearing nuclei. Chromatophores are characteristic of the group and may assume various shapes. A red pigmented oily substance is present in most of the chlorophyll-bearing flagellates as a rod-shaped, oval or discoidal mass called the stigma. Reproduction is usually by longitudinal division. In some genera the cuticle is developed into stalks or collar-like outgrowths. Others produce chitinous shells or masses of jelly and are connected into arborescent or spherical colonies.

ORDER CHRYSOMONADIDA

FAMILY MALLOMONADIDÆ.

Mallomonas. — Free-swimming animalcules, oval or elliptical, persistent in shape; surface covered with overlapping horny plates from which arise long hair like setæ; under low power the surface has a crenulated appearance. One long, slender anterior flagellum; indistinct contractile vacuole. Endoplasm vacuolar, greenish or yellowish. Length from 20 to 40 μ . (Pl. XIII, Fig. 3.)

FAMILY ISOCHRYSIDÆ.

Syncrypta. — Free-swimming animalcules, united into spherical clusters as in Synura, without lorica, but with the entire colony immersed within a gelatinous matrix, beyond the periphery of which the flagella alone project; two subequal flagella; brownish lateral color bands evenly developed; one or two pigment-spots; contractile vacuole between the color-bands. Length of zooids about 10 μ . Diameter of colony about 50 μ , including gelatinous zoöglœa. There is but one species, *S. volvox*. It resembles Synura. It is not common. (Pl. XII, Fig. 11.)

Synura. — Free-swimming animalcules, united in subspherical social clusters, each zooid contained in a separate membranous sheath or lorica, the posterior extremities of which are stalk-like and confluent; two subequal flagella, sometimes long; pigment spots minute or absent; two brown color bands produced equally throughout the length of the two lateral borders; a vacuolar space at the anterior extremity and several contractile vacuoles; oil globules often observed. Length of individual zooids about 35 μ . diameter of clusters varies from 30 to 100 μ . There

is one species, *S. wella*, with several varieties. The colonies move with a brisk rolling motion, caused by the combined action of the flagella. Common in swamp waters. (Pl. XII, Fig. 9.)

FAMILY OCHROMONADIDÆ.

Uroglenopsis. — Animalcules forming almost colorless spheroidal colonies barely visible to the naked eye. The matrix of the colony is a transparent gelatinous shell filled with a watery substance. The zooids are embedded on the periphery, with their flagella extending outward and by their vibration causing the colony to revolve. The zooids are pyriform, with anterior border rounded and truncated, tapering posteriorly and continued backward as a contractile thread; with two light yellowish-green pigment bands; one eyespot at the base of the flagella; two unequal flagella; one or more contractile vacuoles; oil globules and a large amylaceous body often present. Length of zooids is about 6 to 12 μ . The colonies are from 200 to 500 μ in diameter. There are several rather indistinct species. The zooids multiply by division into twos or fours. The colonies also divide, a hollow first appearing on one side, followed by a rounding at the two poles and a subsequent twisting apart. The *Uroglenopsis* colonies are very fragile; *U. americana* is the most common. (Pl. XII, Figs. 12 and 13.)

Dinobryon. — Animalcules with urn- or trumpet-shaped loricae attenuated posteriorly and set one into another so as to form a compound branching polythecium. The zooids are elongate ovate, attached to the bottom of the loricae by transparent elastic threads; two unequal flagella; two brownish or greenish lateral color bands; a conspicuous pigment spot; nucleus and contractile vacuole sub-central. The polythecium is constructed through the successive terminal gemmation of the zooids. Length of separate loricae 15 to 60 μ . The polythecium may contain 2 to 500 loricae. The usual number is between 25 and 50. Reproduction takes place by spore formation. The spores sometimes remain attached to the polythecium, or they may become scattered. When free they are liable to be mistaken for small *Cyclotella*. The spores are from 8 to 20 μ in diameter. There are several species. *D. sertularia* is the most common. (Pl. XIII, Fig. 1.)

ORDER CRYPTOMONADIDA

FAMILY CRYPTOMONADIDÆ.

Chilomonas. — Egg-shaped, anterior end truncated and notched with a well-marked gullet extending to the middle of the body. Upon the upper side of the notch arise two flagella. Colorless with an anterior contractile vacuole and a posterior nucleus. Length about 40 μ . *Chilomonas paramecium* is an extremely common species, occurring in pond water and is especially numerous in infusions.

Cryptomonas. — Free-swimming animalcules, illoricate, but persistent in form, ovate or elongate, compressed asymmetrically; flagella two, long, equal in length, issuing from a deep groove or furrow; large oral aperture at the base of the flagella continued backward as a tubular pharynx; two lateral bright green color bands; conspicuous nucleus and contractile vacuole; oil globules often present. Length from 40 to 60 μ . (Pl. XIII, Fig. 2.)

ORDER DINOFLAGELLIDA

Most of these forms are covered by distinct shells composed of a cellulose-like substance — a few have none. All with the exception of the *Adinina* have characteristic cross and longitudinal furrows. The

surface-dwelling forms generally have distinct chromatophores; some of these and all of the forms living in the depths have none. Flagella are two in number; one, a transverse flagellum, vibrates in the transverse groove or furrow. The other flagellum is more slender and vibrates freely in the water. Because of the presence of the cellulose-like shell, chlorophyll, starch granules and a holophytic mode of nutrition the Dinoflagellates are often classed in the vegetable kingdom. Many are marine forms. Some are phosphorescent. Vacuoles of a somewhat different type from the usual contractile vacuole are always present. Multiplication in the freely moving forms is by simple longitudinal binary fission. Shelled forms have a more complicated division process.

Glenodinium. — Free-swimming animalcules covered with a smooth, cellulose shell not made up of facets, consisting of two subequal parts. There is a conspicuous transverse groove and a much less conspicuous secondary groove. Two typical flagella. Body ovate. Color brownish. Pigment spot sometimes present. Length about 40 to 55 μ . Glenodinium is often surrounded by a wide, irregular mass of jelly. (Pl. XIII, Fig. 7.)

Gymnodinium. — Quite similar to Peridinium, but without a protecting shell.

Peridinium. — Free-swimming animalcules enclosed within a cellulose shell composed of polygonal facets. With a high power the facets exhibit a delicate reticulation. A transverse groove divides the body into two subequal parts. A second groove extends from the first toward the apical extremity. Two flagella, one in the transverse groove, the other proceeding from the junction of the two grooves. Color yellowish-green or brown. There are one or more pigment spots. Length from 40 to 75 μ . There are several species. *P. tabulatum* is the most common. (Pl. XIII, Fig. 5.)

Ceratium. — Free-swimming animalcules enclosed within a shell consisting of two subequal segments, one or both of which are produced into conspicuous horn-like prolongations, often covered with tooth-like processes. There is a central transverse furrow and a second groove extending from the center of the ventral aspect toward the anterior pole. Two flagella, one of which lies in the transverse groove. The brown color is not as marked as in Peridinium. Length from 25 to 150 μ . There are several species, varying considerably in the character of the horn-like projections. (Pl. XIII, Fig. 6.)

ORDER PHYTOMONADIDA

This order includes the most plant-like forms of Protozoa. The chromatophores are colored with chlorophyll sometimes masked by the presence of karotin. The individual monads are small and are never metabolic. Flagella vary from 1 to 2, rarely more. A number of contractile vacuoles are grouped at the anterior end. Reproduction as usual is by longitudinal binary fission. Encystment stages are common.

FAMILY CHLAMYDOMONADIDÆ.

Chlamydomonas. — Animalcules ovate, with two or more flagella, one large green color mass, a delicate membranous shell, usually with a pigment spot and one or more

contractile vacuoles. The protoplasm divides into new individuals within the envelope. Length from 10 to 30 μ . (Pl. XIII, Fig. 4.)

FAMILY VOLVOCIDÆ. — Often included under Protozoa. In this work they are classified as Chlorophyceæ. (See page 470.)

ORDER EUGLENIDA

Somewhat large and highly developed forms with firm, contractile, elastic cortical substance; some forms are stiff, others are capable of annular contraction and worm-like elongation. At the base of the flagellum there is a mouth leading into a pharyngeal tube, near which is a contractile vacuole. The majority of forms have only one flagellum but two are not uncommon.

FAMILY EUGLENIDÆ.

Euglena. — Free-swimming animalcules, fusiform or elongate, exceedingly flexible in form; with highly elastic cuticle terminating posteriorly in a tail-like prolongation; endoplasm bright green or reddish; flagellum flexible, issuing from an anterior notch at the bottom of which is the oral aperture and a red pigment spot. There are several common species. *E. viridis* is the most common. It is often found in immense numbers in stagnant pools, forming a characteristic green or reddish scum. Length varies from 40 to 150 μ . *E. acus* is an elongated form with tapering ends. It is longer than *E. viridis*, but narrower. It is also less variable in form. *E. deses* is a very long cylindrical form. (Pl. XII, Fig. 6.)

Phacus. — Free-swimming animalcules; form persistent, leaf-like, with sharp-pointed, tail-like prolongation; terminal oral aperture and tubular pharynx; flagellum long, vibratile; surface indurated; endoplasm green, with red pigment spot; contractile vacuole large, subspherical. Length about 50 μ , but quite variable. (Pl. XII, Fig. 8.)

Trachelomonas. — Monoflagellate animalcules, changeable in form, enclosed within a free-floating, spheroidal, indurated sheath or lorica; flagellum protruded through an aperture in the lorica. The color of the animalcule is green, with a red pigment spot; the color of the lorica is generally a reddish-brown. There are several species. Diameter of lorica generally about 25 μ . (Pl. XII, Fig. 7.)

ORDER CHLOROMONADIDA

A group of comparatively rare forms with obscure affinities. Vacuole system similar to that of the Euglenida. Contain starch, grass-green chromatophores of discoidal form in chlorophyll-bearing types.

Raphidomonas (Gonyostomum). — Animalcules free-swimming; ovate-elongate, flexible body, widest anteriorly and tapering posteriorly, two to three times as long as wide; two flagella, one of them trailing; oral aperture at anterior end conducts to a conspicuous triangular or lunate pharyngeal chamber; contractile vacuole conspicuous; nucleus ovate; a brownish germ sphere posteriorly located; many large bright green chlorophyll bodies; numerous rod-like bodies called trichocysts; oil globules often present. Length 40 to 70 μ . Reproduction by spores formed in the germ sphere. One species, *R. semen*. The genus *Trentonia*, described by Dr. A. C. Stokes, is similar to *Raphidomonas* except that it has no trichocysts. (Pl. XII, Fig. 5.)

CLASS II. ZOÖMASTIGODA

Animal flagellates with no chromatophores, no chlorophyll and no paramylum granules. Vacuole a simple vesicle. Cortical differentiations less extensive than in the Phytomastigoda. Kinetic and locomotor apparatus more complex than in the plant flagellates. In other respects the animal flagellates are very similar to the plant forms.

ORDER PROTOMASTIGODA

This order includes a great variety of heterogeneous types of flagellates. Flagella chiefly limited to one or two. When two are present they may be equal in length or there may be a well-marked primary and a secondary flagellum. Protoplasmic collars surrounding the bases of the flagella like the collar cells of sponges are present in Choanoflagellidæ and Phalansteriidæ. Colonies are frequently formed. Reproduction is by longitudinal binary fission.

FAMILY CHOANOFLAGELLIDÆ. — Flagellates provided with an upstanding collar surrounding the anterior pole of the cell, from which the single flagellum springs. (Omitted from this work.)

FAMILY MONADIDÆ.

Monas. — Very minute, free-swimming animalcules, colorless, globose or ovate, plastic, with no distinct cuticle; both flagella active, terminal; no distinct mouth. Several species, commonly found in vegetable infusions. Their length varies from 2 to 10 μ . They move with a "swarming" motion. (Pl. XII, Fig. 2.)

Anthophysa. — Animalcules colorless, obliquely pyriform, attached in spherical clusters to the extremities of slightly flexible, granular, opaque, more or less branching pedicles; two flagella, one longer than the other; no distinct mouth. In the common species, *A. vegetans*, the pedicle is dark brown and longitudinally striated. The detached stems somewhat resemble *Crenothrix* when observed with a low power. Zooids about 5 μ long; clusters 25 μ in diameter. Common in swamp water. (Pl. XII, Fig. 3.)

FAMILY BODONIDÆ.

Cercomonas. — Animalcules free-swimming, ovate or elongate, plastic, with a single long flagellum at anterior extremity and a caudal filament at the opposite extremity; no oral aperture. There are several species. Their length varies from 10 to 25 μ . (Pl. XII, Fig. 1.)

SUB-PHYLUM INFUSORIA

In its broadest sense the word Infusoria includes all the Protozoa except the Rhizopoda and Sporozoa. As used here, following Bütschli, it includes only the classes Ciliata and Suctoria.

CLASS CILIATA

Protozoa of relatively large size, furnished with cilia, but not with flagella. The cilia occur as a single band surrounding the oral aperture or are dispersed over the entire body. Modification of the cilia into setae or styles is sometimes observed. There is generally a well-developed oral and anal aperture. The nucleus varies in different genera. Besides one larger, oblong nucleus (macronucleus) a smaller one (micronucleus) is often present. One or more contractile vacuoles are present. They all possess a delicate but well-defined ectoderm, elastic, but constant in form. They occur naked or enclosed in horny or siliceous shells or in gelatinous envelopes. Some genera are stalked. Multiplication takes place by transverse fission. Conjugation has been observed in many forms. According to some authorities this has to do with the restoration of the vitality of the organisms. According to others it furnishes the basis of variation due to biparental inheritance. Many of the Ciliata are parasites in higher animals.

The Ciliata are divided into five orders according to the character and distribution of their cilia.

ORDER HOLOTRICHIDA

Ciliata with but one sort of cilia, these covering the body uniformly and almost completely. A variously modified extensile or undulating membrane sometimes present. Oral and anal orifices usually conspicuous. Trichocysts sometimes present in the cuticular layer.

Paramecium. — Animalcules free-swimming, ovate or elongate, asymmetrical, more or less flexible, but persistent in shape. Finely ciliated throughout the cilia of the oral region, not differing in size or character from those of the general surface of the body. An oblique groove developed on the ventral surface, at the posterior extremity of which is situated the oral aperture. Cortical layer usually enclosing trichocysts. Contractile vesicles and nucleus conspicuous, the former sometimes stellate. There are several species. The most important is *P. aurelia*, which is often found in sewage-polluted and stagnant water. It is colorless, has a length of about 225 μ , and moves with a brisk rotatory motion. (Pl. XIV, Fig. 4.)

Nassula. — Animalcules ovate, cylindrical, flexible, but not polymorphic, usually highly colored — rose, red, blue, yellow, etc. Oral aperture lateral. Pharynx armed with a simple horny tube or with a cylindrical fascicle of rod-like teeth. Entire surface of cuticle finely and evenly ciliate. The cortical layer sometimes containing trichocysts. There are several species, varying in color, shape, and size. Length 50 to 250 μ . (Pl. XIV, Fig. 5.)

Coleps. — Animalcules ovate, cylindrical, or barrel-shaped, persistent in shape, cuticular surface divided longitudinally and transversely by furrows into quadrangular facets; these facets are smooth and indurated, the narrow furrows soft and clothed with cilia; the anterior margin mucronate or denticulate; the posterior extremity mucronate and provided with spines or cusps. Oral aperture apical,

terminal, surrounded with cilia. Anal aperture at posterior extremity. Color gray or light brown. The most common species is *C. hirtus*, which has a length of about 60 μ . (Pl. XIV, Fig. 6.)

Enchelys. — Animalcules free-swimming, elastic, and changeable in shape, pyriform or globose. Oral aperture situated at the termination of the narrower, and usually oblique, truncate anterior extremity. Anal aperture at the posterior termination. Cuticular surface finely and entirely ciliate; the cilia are longer in the region of the mouth. Few species. Length about 25 to 50 μ . (Pl. XIV, Fig. 7.)

Trachelocerca. — Animalcules colorless, highly elastic, and changeable in form, the anterior portion produced as a long, flexible, narrow, neck-like process, the apical termination of which is separated by an annular constriction from the preceding part, and is perforated apically by the oral aperture. Cuticular surface evenly and finely ciliate; a circle of larger cilia developed around the oral region. Length of extended body about 150 μ . Few species. (Pl. XIV, Fig. 8.)

Pleuronema. — Animalcules ovate, colorless. Oral aperture situated in a depressed area near the center of the ventral surface, supplemented by an extensile, hood-shaped, transparent membrane or velum, which is let down or retracted at will. Numerous longer vibratile cilia stationed at the entrance of the oral cavity. The general surface of the body clothed with long, stiff, hair-like setæ. The cortical layer usually containing trichocysts. Length 60 to 100 μ . Few species. (Pl. XIV, Fig. 9.)

Colpidium. — Animalcules free-swimming, colorless, kidney-shaped. Entirely ciliate. Oral aperture inferior, subterminal. Pharynx supported throughout its length by an undulating membrane which projects exteriorly in a tongue-like manner. Two nuclei, rounded, sub-central. Length 50 to 100 μ . One species. (Pl. XV, Fig. 1.)

ORDER HETEROTRICHIDA

Ciliata possessing two distinct systems of cilia, one a band or spiral or circlet of long cilia developed in the oral region, the other composed of short, fine cilia covering the entire body. The cortical layer is usually highly differentiated.

Stentor. — Animalcules sedentary or free-swimming at will; bodies highly elastic and variable in form; when swimming and contracted, clavate, pyriform, or turbinate; when fixed and extended, trumpet-shaped, broadly expanded anteriorly, tapering off and attenuated toward the attached posterior extremity. Peristome describing an almost complete circuit around the expanded anterior border, its left-hand extremity or limb spirally involute, forming a small pocket-shaped fossa conducting to the oral aperture, the right-hand limb free and usually raised considerably above the opposite or left-hand one. Peristomal cilia cirrose, very large and strong; cilia of the cuticular surface very fine, distributed in even longitudinal rows, occasionally supplemented by scattered hair-like setæ. Nucleus band-like, moniliform, or rounded. Contractile vesicle complex. Multiplication by oblique fission and by germs separated from the band-like endoplast. There are many species, some of large size, colorless, or greenish, bluish, brownish, etc. (Pl. XIV, Fig. 2.)

Bursaria. — Animalcules free-swimming, broadly ovate, somewhat flattened on one side, anteriorly truncate. Peristome field pocket-shaped, deeply excavate, situated obliquely on the anterior half of the body, having a broad oral fossa in front, and a cleft-like lateral fissure, which extends from the left corner of the contour

border to the middle of the ventral side; no tremulous flap. Pharynx long, funicular, bent toward the left, and forming a continuation of the peristome excavation. Adoral ciliary wreath broad, much concealed, lying completely within the peristome cleft. Cuticular cilia fine, in longitudinal rows. Anus posteriorly situated, terminal. Nucleus band-like, curved, or sinuous. Contractile vesicles distinct, usually multiple. Few species. Length 300 to 500 μ . (Pl. XIV, Fig. 3.)

ORDER OLIGOTRICHIDA

Organisms with greatly reduced cilia or none at all. Motile organs composed of membranelles. Adoral zone forming a ring (complete or nearly so) around the margin of the peristome. Peristome usually at right angles to the long axis of the body.

Halteria. — Animalcules free-swimming, colorless, more or less globose, terminating posteriorly in a rounded point. Oral aperture terminal, eccentric, associated with a spiral or subcircular wreath of large cirrose cilia. A zone of long hair-like setæ or springing-hairs developed around the equatorial region, the sudden flexure of which appendages enables the organism to progress through the water by a series of leaping movements, in addition to their ordinary swimming motions. Length 15 to 30 μ . There are several species, some of them colored green. (Pl. XIII, Fig. 9.)

Tintinnus. — Animalcules ovate or pyriform, attached posteriorly by a slender retractile pedicle within an indurated sheath or lorica. The shape of the lorica is generally cylindrical; it is free-floating; it is somewhat mucilaginous and attracts to its outer surface foreign particles, such as grains of inorganic matter, diatom shells, etc. The peristome field of the organism occupies the entire anterior border, circumscribed by a more or less complex circular or spiral wreath of long, powerful, cirrose cilia, the left limb or extremity of which is spirally involute and forms the entrance to the oral fossa. This fossa is continued as a short, tubular pharynx. Anus posteriorly situated, subterminal. Cuticular cilia very fine, distributed evenly throughout, clothing both the body and the retractile pedicle. Length of lorica 80 to 150 μ . There are many species, varying greatly in the size and shape of the loricae. In the fresh-water forms the lorica is generally cylindrical. Another genus, *Tintinnidium*, varies from *Tintinnus* only in having a more mucilaginous sheath and in being permanently attached to foreign objects. (Pl. XIII, Fig. 12.)

Codonella. — Animalcules conical or trumpet-shaped, solitary, free-swimming, highly contractile, inhabiting a helmet- or bell-shaped lorica, to which they are attached by their posterior extremity. The anterior region truncate or excavate, forming a circular peristome having an outer fringe of about twenty long, tentacle-like cilia, and an inner collar-like border, or frill, which bears an equal number of slender, lappet-like appendages. Entire cuticular surface clothed with fine, vibratile cilia. Lorica not perforated, of chitinous consistence, often of a brown color, sometimes sculptured or mixed with foreign granular substances. Length of lorica 50 to 150 μ . Several species, mostly marine. (Pl. XIV, Fig. 1.)

ORDER HYPOTRICHIDA

Ciliata in which the body is flattened and the locomotive cilia are confined to the ventral surface, and are often modified and enlarged to the condition of muscular appendages. Usually an adoral band of

cilia, like that of *Heterotrichida*. Dorsal surface smooth or provided with tactile hairs only. Mouth and anus conspicuous.

Euplotes. — Animalcules free-swimming, encuirassed, elliptical or orbicular, with sharp laminate marginal edges, and usually a plane ventral, and convex, sometimes furrowed, dorsal surface. Peristome field arcuate, extending backward from the frontal border to or beyond the center of the ventral surface, sometimes with a reflected and ciliate inner border. Frontal styles six or seven in number; three or more irregularly scattered ventral styles, and five anal styles; four isolated caudal styles along the posterior margin. Endoplast linear. Single spherical contractile vesicle near anal aperture. Length about 125 μ . (Pl. XIII, Fig. 8.)

ORDER PERITRICHIDA

Ciliata with the cilia arranged in one anterior circle or in two, an anterior and a posterior; the general surface of the body destitute of cilia. The Peritrichida are sometimes divided into two suborders, the free-swimming forms and the attached forms.

Vorticella. — Animalcules ovate, spheroidal, or campanulate, attached posteriorly by a simple undivided, elongate and contractile, thread-like pedicle; the pedicle enclosing an elastic, spirally disposed, muscular fibrilla, and assuming suddenly on contraction a much-shortened and usually corkscrew-like contour. Adoral system consisting of a spirally convolute ciliary wreath, the right limb of which descends into the oral cleft, the left one obliquely elevated and encircling the ciliary disk. The entire adoral wreath contained within and bounded by a more or less distinctly raised border — the peristome — between which and the elevated ciliary disk, on the ventral side, the widely excavated cleft or vestibulum is situated. The vestibulum is continued further into a conspicuous cleft-like pharynx, and terminates in a narrow tubular oesophagus. Anal aperture opening into the vestibulum. Contractile vesicle single, spherical, near the vestibulum. Nucleus elongate. Multiplication by longitudinal fission, by gemmation, and by the development of germs. There exists a very large number of species, varying considerably in size and shape. The length varies from 25 to 200 μ . *Vorticella* is often found floating in water attached to masses of *Anabaena*, etc. (Pl. XIII, Fig. 10.)

Zoëthamnium. — Animalcules structurally identical with those of *Vorticella*, ovate, pyriform, or globular, often dissimilar in shape and two sizes, stationed at the extremities of a branching, highly contractile pedicle or zoödendrium. Numerous species.

Carchesium. — Forming richly branched colonies in which the stalk muscles of the single individual are not attached, but end abruptly at the base of the individual stalks, enabling the individuals to contract independently. Animals all alike in size and structure. Ciliated spiral forms about one and a half circles. Macronucleus horseshoe-shaped, micronucleus close to the macronucleus. A single contractile vacuole. Colonies of *Carchesium* sometimes attain a diameter of 1 cm. These are found attached to stems of plants, stones, etc., in fresh water.

Epistylis. — Animalcules campanulate, ovate, or pyriform, structurally similar to *Vorticella*, attached in numbers to a rigid, uncontractile, branching, tree-like pedicle or zoödendrium; the zoöids usually of similar size and shape. Numerous species. (Pl. XIII, Fig. 11.)

CLASS SUCTORIA

Protozoa with neither flagellate appendages nor cilia in their adult state, but seizing their food and effecting locomotion, when unattached, by means of tentacles. These are simply adhesive or tubular and provided at their distal extremity with a cup-like sucking-disk. Nucleus usually much branched. One or more contractile vesicles. Multiplication by longitudinal or transverse fission or by external or internal bud-formation. The young forms are ciliated. Most of the Suctoria are sedentary.

Acineta. — Animalcules solitary, ovate or elongate, secreting a protective lorica, to the sides of which they are adherent or within which they may remain freely suspended. Lorica transparent, triangular or urn-shaped, supported upon a rigid pedicle. Tentacles suctorial, capitate, distributed irregularly or in groups. There are many species. (Pl. XV, Fig. 2.)

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CHAPTER XXVII

ROTIFERA

The Rotifera, or Rotatoria, comprise a well-defined group of multicellular animals. They are considered by most authorities to have arisen as a degenerate off-shoot of the Annelid worms, and are ordinarily regarded as a class of the heterogeneous phylum Vermes, the Lower Worms.

There are at present about 950 known species, with new ones being added to the list as the interest in the group becomes more and more widespread. Only about 300 species have been identified in this country, but this is to be taken as indicative of lack of investigation rather than of small numbers of native forms.

The majority of the Rotifera are solitary, motile, free-living animals, although some form colonies; quite a number are sessile in the adult condition, and a very few are parasitic. They are of cosmopolitan distribution and are found in all sorts of habitats. They are almost entirely confined to fresh water, but a few are marine. There are 50 species in the arctic regions, and many in the tropics. They sometimes occur in large numbers, but never to such an extent as to cause trouble in reservoirs and water supplies as do certain of the protozoa and algæ.

Anatomy and Physiology. — The rotifera are quite highly organized, although the fact that many of them are smaller than some of the larger protozoa led early investigators to consider them of equally simple structure. They have a well-defined digestive system, including a buccal funnel; a "mastax," or masticatory organ peculiar to the group; digestive glands; a short œsophagus; a stomach; an intestine; and a cloaca. In some forms, i.e. Asplanchna, the intestine is absent and the digestive tract is a "blind alley." Into the cloaca also empties the excretory system, which consists of a single pair of lateral canals containing "flame-cells," and a median contractile vesicle usually placed quite near to the cloaca. The reproductive system is very conspicuous. There is a single ovary, containing about eight oöcytes, and a large vitellarium or yolk-tissue. A duct can be demonstrated in some forms, leading from the ovary to the cloaca, and in some cases the eggs hatch in an expanded portion of this duct and the young are extruded alive from the cloaca. There are two kinds of muscles, those of the body wall being typically striated. The nervous system consists of a lobed

cerebral ganglion, or "brain," from which run fibers to the various organs. From one to three eyes are usually to be found embedded in the anterior part of the "brain." Other sense organs, chiefly tactile, are sometimes present.

The organisms are protected by a firm, homogeneous cuticle, which in some forms is thrown into segmental folds by the manner in which the muscles are attached beneath it, giving a pseudo-metameric appearance to the animal. In one order there is an additional gelatinous sheath which is thrown off from the skin in rings as the animal develops, and in another sub-order, the cuticle is hardened by deposits of a chitinous nature into a firm shell of one or two valves, called a "lorica."

Certain of the sessile forms have still other kinds of protective sheaths; one genus, *Melicerta* (Pl. XV, Fig. 4), building up a tube of pellets made by the animal itself from particles, collected from the surrounding water — the resultant structure being a sort of chimney into which the animal recedes when disturbed.

The rotifera are all bilaterally symmetrical, with a dorsal and a ventral surface, and with definite head and tail regions. The majority of them are broadest anteriorly and taper posteriorly to a "foot" of some form.

In the majority of the genera of the rotifera, only the females have been seen, the eggs developing parthenogenetically, and the males, in those genera which possess them, appearing only toward the approach of winter, when the fertilized "winter," or resting eggs are produced. Male rotifers are highly specialized for their work of reproduction. They are utterly lacking in digestive organs, and their lives are necessarily of short duration.

Three features of the rotifera deserve special attention. They are peculiar to this group, and form the basis for the classification of its members. They are the "mastax," the "foot," and the "corona."*

The "mastax" is a sort of muscular sac, forming a part of the pharynx, and having an opening into the mouth anteriorly and into the short oesophagus posteriorly. In it are contained the "trophi," a number of interacting, calcareous structures, moved by the muscles of the mastax, whose function it is to tear and crush the food of the animal as it streams through the mastax on its way to the stomach.

The mastax is almost continually in action, even when the animal is not actually feeding, which has led some to ascribe a respiratory function to it also. It begins to pulsate even before the egg is hatched, and was thought by early observers to be the "heart" of the animal!

As a means of readily determining whether a minute animal is a

* These features are illustrated in Figs. A to P, Plate XVI.

rotifer or a protozoön, the characteristic pulsing of the mastax should be looked for, and is a certain criterion.

The "foot," or posterior extremity of the body, may be one of several types, or may be entirely lacking, or modified into other organs. It may be soft and flexible, or rigid, with or without joints. It may be retractile or non-retractile. It may terminate in an adhesive disk, or in a ciliated expansion, or it may be divided into two toes. The pseudometamerism referred to above, when present, is generally confined to the foot region, and the divisions of the foot can then usually be telescoped into one another by the contraction of the longitudinal muscles. Almost without exception, the foot contains glands which secrete an adhesive mucus, enabling the animal to attach itself at will to the substratum, while jerking the body to and fro in the water in search of food.

The "corona," or disk bearing the ciliary wreath, is borne upon the head and surrounds the mouth and sense organs. In the primitive and simple forms the ciliary wreath is complete and unbroken, while in the higher types the corona is specialized and consists of a series of lobes, prominences, projections, etc. bearing groups of cilia. The corona is usually edged by a double row of cilia, the anteriormost row being known as the "trochum," the posteriormost as the "cingulum." The corona is lacking in the few forms that are parasitic.

The activity of the corona produces currents in the water which, if the animal is unattached, effect its locomotion, or if the animal is attached permanently or temporarily, bring food particles toward it and sweep them down the buccal funnel.

The beating of the cilia occurs in such a peculiar way that an optical illusion is sometimes produced in which the cilia seem to be moving in a regular procession around the rim of the trochal disk. This wheel-like appearance of the corona is responsible for the name of "Wheel-animalcules," which was given to the group by the earliest observers, who thought they could see the wheels, with their axles and all, and so drew them.

Peculiarities of certain of the rotifera could be enumerated without end. They are among the most fascinating of subjects for microscopic study.

Members of one family, the Philodinidæ, are extraordinarily resistant to desiccation, and their dried bodies may be recovered and revived from gutter dust, mosses, and many places where the existence of such organisms would not ordinarily be suspected. This property of "suspended animation" over a period of months or even years was formerly attributed promiscuously to all rotifera alike, but has recently been found to be restricted to members of the above-mentioned family.

KEY TO THE FAMILIES DESCRIBED

1. Animals sessile.	4
1. Animals free-swimming.	2
2. Animals swimming with their ciliary wreath and creeping like a leech.	Philodinidae
2. Animals swimming with their ciliary wreath and creeping with their toes	3
3. Animals with a lorica.	7
3. Animals without a lorica.	5
4. Corona with prominent non-vibratile cilia usually on lobes; vibratile cilia very small.	
	Floscularidae
4. Corona without non-vibratile cilia; colonial or not.	Melicertidae
5. Foot with two toes present; auricles often present.	Notommatidae
5. No foot present.	6
6. No long lateral appendages.	Asplanchnidae
6. Long lateral appendages present, with which the animal jumps.	Triarthridae
7. Foot absent.	Anuraeidae
7. Foot present.	8
8. Foot transversely wrinkled or ringed, but not jointed.	9
8. Foot not transversely wrinkled or ringed, often jointed, with 1 or 2 toes.	10
9. Foot ending in a ciliated cup, without toes.	Pterodinidae
9. Foot ending in two toes.	Brachionidae
10. Foot usually long and jointed, toes very long; lorica produced dorsally into a head shield.	
	Dinocharidae
10. Foot usually very short with 1 or 2 slender and usually long toes; lorica flattened and ovate.	11
11. An arched shield over the head.	Coluridae
11. No arched shield over the head.	12
12. Toes 1 or 2 in number, slender and rod-shaped.	Cathypnidae
12. Toes 2 in number, long and blade-shaped, often diverging.	Euchlanidae

CLASSIFICATION AND DESCRIPTION

The rotifera have recently been renamed and reclassified by Haring (1913) but as the old names and classification according to Hudson and Gosse are still in almost universal use, it has seemed advisable to retain them in this work. The following brief classification and descriptions of the common genera are taken from Hudson and Gosse:

ORDER RHIZOTA

Rotifera fixed when adult; usually inhabiting a gelatinous tube thrown off from the skin. Foot transversely wrinkled, not contractile within the body, ending in an adhesive disk; without telescopic joints, never furcate.

FAMILY FLOSCULARIDÆ. — Corona produced into lobes, bearing long, non-vibratile cilia. Vibratile cilia few, short, and confined to a single half-circle about the central mouth. -

Floscularia. — Coronal lobes blunt, expanded, or absent. Very long, non-vibratile cilia. Foot very long, ending in an adhesive disk. Inhabits a very thick gelatinous tube, into which the animal recedes when disturbed. The young of all species are free-swimming. 30 species. (Pl. XV, Fig. 3.)

FAMILY MELICERTIDÆ. — Usually colonial and usually tubiculous. No non-vibratile cilia. Corona membranous, two- or four-lobed. Ciliary wreath double.

Melicerta. — Corona four-lobed. Inhabiting tubes built up of spherical or conical pellets, closely packed or loosely held in general gelatine. Fairly common attached to lily-pads, etc. (Pl. XV, Fig. 4.)

Limnias. — Corona two-lobed. Inhabiting thin, homogeneous tubes, sometimes annulated. Very common on duck weed rootlets and the like.

Conochilus. — Corona horseshoe-shaped, transverse. Forming free-swimming colonies of several individuals inhabiting coalescent gelatinous tubes. Fairly common. (Pl. XV, Fig. 5.)

ORDER BDELLOIDA

Rotifera that swim with their corona and creep like a leech. Foot retractile, telescopic, usually with 3 toes and 2 or 4 spurs near its extremity.

FAMILY PHILODINIDÆ. — Corona a pair of circular, retractile lobes. A proboscis, bearing a few cilia, present. Proboscis thrown back like a cape when corona is everted. Dorsal antenna very conspicuous.

Rotifer. — Two red eyes on the proboscis. Body long and slender. Common among mosses and dust, as well as in water. (Pl. XV, Fig. 6.)

Philodina. Two red eyes on the neck. Body usually stout and often colored. Extremely common among plants, dust, and in water.

ORDER PLOIMA

Rotifera that swim with their corona and attach themselves temporarily by the foot. (The largest and most important order.)

SUB-ORDER ILLORICATA

Cuticle flexible. No lorica. Foot, when present, almost invariably furcate.

FAMILY ASPLANCHNIDÆ. — Large, transparent, and sac-shaped. Intestine and cloaca absent.

Asplanchna. — Foot absent. Trophi large and not in a mastax. Viviparous. Very large and transparent. (Pl. XV, Fig. 8.)

FAMILY TRIARTHRIDÆ. — Foot absent. Paired lateral appendages by means of which the animal skips through the water.

Polyarthra. — Body rectangular, with six serrated spines attached to each shoulder. Eggs carried attached to the body after extrusion. One species, *P. platyptera*. Two varieties, major and minor. (Pl. XVI, Fig. 2.)

FAMILY NOTOMMATIDÆ. — Foot furcate. Body soft and elongate; corona oblique, covered with clusters of cilia and often with a pair of lateral ciliated projections called "auricles." This family is the most typical, the most highly organized, of the Rotifera.

Diglena. — Body subcylindrical. Two eyes. Foot furcate. Trophi protrusible for the purpose of cropping vegetation, etc. (Pl. XVI, Fig. 4.)

Furcularia. — Body cylindrical. One eye. Foot furcate, with toes often much longer than body, but not usually of equal length. Fairly common.

SUB-ORDER LORICATA

Cuticle stiffened into a lorica, which is usually flattened. Foot various.

FAMILY DINOCHARIDÆ. — Foot very long with two long toes. Lorica heavy, more or less cylindrical and usually produced dorsally into a sort of shield above the head.

Dinocharis. — Lorica tessellated and transversely ridged. Foot jointed, one joint bearing two spurs. Toes divergent. Fairly common.

FAMILY EUCHLANIDÆ. — Large and transparent. Lorica with a convex dorsal and a flat ventral valve. Foot jointed, with two blade-shaped and divergent toes.

Euchlanis. — Lorica usually oval and flat, but sometimes triangular in end-view. One eye. Very common.

FAMILY CATHYPNIDÆ. — Large and transparent. Lorica with a convex dorsal and a flat ventral valve, the two being separated by a deep groove on each side. Foot very short, with one or two rod-shaped toes.

Monostyla. — Body oval or nearly circular, with one rod-like toe. Very common among aquatic plants. Often creeps on submerged stems, cropping with its protrusible trophi.

FAMILY COLURIDÆ. — Lorica consisting usually of a single dorsal valve, produced into a head shield.

Colurus. — Body sub-globose, more or less compressed. Lorica of two lateral plates, open in front, gaping behind. Frontal hood in form of a non-retractile hook. Foot prominently extruded, of distinct joints, and with two toes. Many species.

FAMILY PTERODINIDÆ. — Lorica very much flattened. Foot long, annulated, and ending in a ciliated cup without toes.

Pterodina. — Lorica almost perfectly circular and extremely flattened. Body wholly retractile within the lorica. Foot issues from opening in lorica in posterior-ventral region. Intestine continues through foot, anus being at the end.

FAMILY BRACHTONIDÆ. — Lorica flattened and open at both ends. Foot long, cylindrical, annulated, and very flexible. Two very short toes.

Brachionus. — Lorica arched dorsally, flattened ventrally, with several spines on anterior margin and sometimes on posterior margin also. Foot very flexible, uniformly annulated but not jointed. Two small toes. One eye. Eggs carried after extrusion. Many species, some marine. Very common. (Pl. XVII, Fig. 1.)

Notus. — Lorica oval and faceted, with two anterior and two posterior spines. Foot jointed, with fairly long toes. No eye.

FAMILY ANURÆIDÆ. — Lorica box-like, broadly open in front, open behind only in a narrow slit. Usually with 6 spines on anterior margin and 1 or 2 on posterior. Foot absent.

Anuræa. Lorica thick-walled and opaque, with ridges on the dorsal surface marking it off into polygonal areas. Eggs carried after extrusion. The empty lorica is seen more often than the living animal. (Pl. XVII, Figs. 2 and 3.)

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CHAPTER XXVIII

CRUSTACEA

The Crustacea belong to the Arthropoda, that is, to that group of segmented animals which have jointed appendages. Most of the larger crustacea are marine; of the smaller forms many are found in fresh water. The organisms vary in size; some are just visible to the naked eye while others are several centimeters in length. The vast majority are the smaller forms measuring from one to three millimeters in length.

General Classification. — The Crustacea are divided into two large groups, the Entomostraca or the smaller forms, and the Malacostraca or larger forms. The latter group of comparatively conspicuous animals is further divided into three orders: the Decapoda, Isopoda, and Amphipoda. The Decapoda include the crayfishes or “crawfish.” They are animals that look like small lobsters. The Isopoda, commonly called “water lice” and “sow bugs,” are flattened from top to bottom and are about a centimeter long. *Asellus communis* is the most widely distributed species. The Amphipoda or “scuds,” are animals about the size of the isopods but can be distinguished from the latter because they are flattened from side to side. The common amphipods belong to the genus *Gammarus*.

The fresh-water Entomostraca are the most commonly found fresh-water crustaceans. They are divided into four orders all of which possess certain similar features. The orders are: the Phyllopoda or fairy shrimps, the Cladocera or water fleas, the Copepoda, and the Ostracoda.

Anatomy and Physiology of the Entomostraca. — The bodies of some of the Entomostraca are segmented and are contained in a horny, leathery, or brittle shell. This shell or carapace is composed of chitin impregnated with variable amounts of carbonate of lime. It is often transparent, and may be striated, reticulated, notched, spinous, etc., in the different genera. The carapace is either composed of two parts (bivalve), similar to the shell of a clam, or it is merely folded and so looks like a bivalve. The carapace may enclose the entire animal, all of the body except the head, or may be segmented like a lobster's shell. Practically all of the Entomostraca possess a pair of large antennæ that are used in swimming. The legs vary in number, position and character. In some genera they are flattened, and have breathing-

plates attached to them, in others they are much branched and are used in swimming or in causing a current of water to pass through the carapace. Most forms possess one conspicuous eye, usually black or reddish, situated in the head region. The animals are well-developed and possess most of the organs of higher animals. There is a simple sac-like heart which causes the circulation of the colorless blood; a well marked digestive tract; muscular, nervous and reproductive organs. The eggs during development may be carried by the female in either a brood chamber or in egg sacs, or they may be laid in packets or singly on leaves, sticks, etc. In some forms (Cladocera) the eggs hatch as fully formed young, which look like the adult, but are smaller in size. In others the young hatch in an immature undeveloped stage, termed the "nauplius" stage. The nauplius can be described as a free-living developmental form that looks unlike the adult. It usually is unsegmented and has but three or four pairs of appendages. After several moults the nauplius becomes adult. All Crustacea grow by shedding the carapace and then rapidly increasing in size. Other habits and features of structure will be considered under the separate orders.

CLASSIFICATION AND DESCRIPTION OF THE ENTOMOSTRACA

ORDER PHYLLOPODA

Body with or without shell. Legs 11 to 60 pairs. Joints foliaceous or branchiform, chiefly adapted for respiration but used also for swimming. One or two pairs of antennæ, neither adapted for swimming. Young hatched in nauplius stage. Animals are graceful swimmers and swim on their backs with the ventral side uppermost.

Eubranchipus (formerly called *Branchipus*). — Body elongate and without shell. Eleven pairs of legs. Antennæ two pairs, the lower horn-like and with prehensile appendages in the male. Color reddish. Two eyes which are stalked and movable. Length about two centimeters. Usually found in spring of year shortly after the snow and ice melt. (Pl. XVIII, Fig. 3.)

ORDER CLADOCERA

Body, except for head, covered with a bivalve carapace. Five or six pairs of leaf-like feet provided with branchiæ, or breathing plates. Usually a large movable compound eye and often a pigment spot inside the head. Second pair of antennæ large, biramous, and used in swimming. First antennæ generally small, except in the males of some species. Heart easily seen in dorsal neck region through the transparent carapace. Intestine conspicuous and usually greenish in color due to algæ that have been eaten. Summer eggs, which are greenish

and many in number, carried in brood chamber between back of animal and shell. The winter eggs, one or two in number, dark in color and encased in a thick-walled, reticulated, roughly triangular or saddle-shaped case. These cases may be found floating on the surface of the water or attached to sticks, grass, etc. Young are released fully formed (no nauplius stage). Five genera are frequently found.

Sida. — Six pairs of similar legs. Body long and narrow. Carapace very transparent. Second antennæ large with dorsal ramus three-jointed and ventral ramus two-jointed. First antennæ small but movable and attached to sides of beak. Intestine nearly straight, at least not coiled. Animals are strong swimmers but frequently adhere to sides of container or to aquatic vegetation by means of a cervical gland. (Pl. XVIII, Fig. 1.)

Daphnia. — Head produced into a beak which bears the terminal bristles of the immovable first antennæ. Second antennæ large with dorsal ramus three-jointed, ventral ramus four-jointed. Valves of carapace oval, reticulated, and terminating caudally in a long spine. Intestine not coiled but bent ventrally at anterior and posterior ends. These animals are usually more abundant in the spring and fall, often occurring in swarms or bunches. In a culture bottle they are constantly in motion never reaching either the surface or the bottom but executing "place maintaining movements." (Pl. XVII, Fig. 9.)

Simocephalus. — Body large, heavy; shell thick and brownish or yellowish in color. Head small, pigment spot prominent. Valves large and subquadrate, varying slightly in shape with age of animal, striate. First antennæ small but movable and bearing one basal spine in female and two in male. Intestine not coiled. These animals occur throughout the year but are more abundant in the spring. They swim rapidly and without noticeable jerks. They either rest on the bottom or remain insecurely attached to the sides of the aquarium by a gland located in the cervical region.

Bosmina. — Head terminated in front by a sharp beak directed forward and down and from the end of which the first antennæ project as long, many-jointed, curved, proboscis-like structures. Swimming-antennæ with one ramus three-jointed and the other four-jointed. Shell oval, yellowish and with a short spine at the lower posterior angle. Pigment spot absent. These animals, in an aquarium, have a tendency to float attached to the surface film as though slightly oily. (Pl. XVII, Fig. 10.)

Chydorus. — Small spherical forms with head prolonged into a sharp rostrum. The pigment spot is large and situated in front and below the compound eye. Swimming-antennæ short. Intestine coiled. Color light yellow to dark brown. Move with an unsteady rolling motion. Very widely distributed. (Pl. XVIII, Fig. 2.)

ORDER COPEPODA

Shell jointed, forming a more or less cylindrical buckler, or carapace, enclosing the head and thorax. The anterior part of the body is composed of ten segments more or less fused. The five constituting the head bear respectively a pair of jointed antennæ, a pair of branched antennules, a pair of mandibles or masticatory organs, a pair of maxillæ, and a pair of foot jaws. The five thoracic segments bear five pairs

of jointed swimming-feet, the fifth pair often rudimentary. There are about five abdominal segments, nearly devoid of appendages, and continued posteriorly by two tail-like stylets. The eggs are carried in either one or two egg sacs, which are posterior in position. Young are hatched in the nauplius stage.

The Copepoda are frequently brightly colored in reds, blues and purples either due to oil globules, or to the integument. Hardly any body of water, which is relatively quiet, is without a population of copepods. They lead a roving, predatory life and well deserve the name of "scavengers." There are many genera and species; the following three genera are quite common.

Cyclops. — Head hardly distinguishable from the body. The thorax and abdomen generally distinguishable, the former having four and the latter six segments. Two pairs of antennæ, the first large and many-jointed and the second smaller and furnished with short setæ; both first antennæ of the male have swollen joints. Two pairs of vigorous, branched foot jaws. One eye, large, and central in position. Two egg sacs. There are many species. (Pl. XVII, Fig. 5.)

Diaptomus. — Resembles Cyclops in general appearance. Thorax and abdomen each with five segments. Antennæ very long, many-jointed, with setæ; the right antenna, only, swollen in the male. Antennules large, bifid, the two unequal branches arising from a common foot stalk. Three pairs of unbranched foot jaws. One egg sac. (Pl. XVII, Fig. 6.)

Canthocamptus. — Resembles Cyclops. The ten segments of the thorax and abdomen not distinguishable. The segments decrease in size as they descend. At the junction of the fourth and fifth segments the body is very movable. Antennæ very short, having but eight segments. Five pairs of swimming feet, much longer than in Cyclops. One egg sac. The animals belonging to this genus are very small and worm-like. Very common. (Pl. XVII, Fig. 7.)

ORDER OSTRACODA

Entire body enclosed in a bivalve shell which is hinged dorsally. One to three pairs of feet. No external ovary. Eggs are laid on external objects and the young hatch in a nauplius stage. Color of shell quite different in different genera and often seems to agree with surroundings.

Cypris. — Shell oval or reniform. First antennæ seven-jointed, with long feathery filaments arising from the last three segments. Second antennæ leg-like, with claws and setæ at the end. Two pairs of feet. Eye single. Color often greenish, brownish or whitish. There are many species. (Pl. XVII, Fig. 8.)

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CHAPTER XXIX

BRYOZOA

The Bryozoa, or Polyzoa, are minute animals forming moss-like or coral-like calcareous or chitinous aggregations. The colonies are called corms, polyzoaria, or ccenœcia. They often attain an enormous size. In the adult stage they lead a sedentary life attached to some submerged object. The animals themselves are small, but easily visible to the naked eye. Some of them are covered with a secreted coating, or sheath, that takes the form of a narrow, brown-colored tube; others are embedded in a mass of jelly. The genera that live in the brown, horny tubes form tree-like growths that often attain considerable length. The branches are sometimes an inch long, and each one is the home of an individual polyzoön, or polypid. The branches, or hollow twigs, are separated from the main stalk by partitions, so that, to a certain extent, each polypid lives a separate existence in its own little case, though each was formed from its next lower neighbor by a process of budding.

Anatomy and Physiology. — The body of the organism is a transparent membranous sac, immersed in the jelly or concealed in the brown opaque sheath. It contains a U-shaped alimentary canal, with a contractile cesophagus, a stomach, and an intestine; a muscular system that permits some motion within the case, and that enables the animal to protrude itself from the case and to extend and contract its tentacles; mesenteries in the form of fibrous bands; an ovary; and a rudimentary nervous system. The organisms possess no heart and no blood-vessels of any kind.

The most conspicuous part of the animal is the circlet of ciliated tentacles. They are mounted on a sort of platform, or disk, called a lophophore, at the forward end of the body. This lophophore, with its crown of tentacles, may be protruded from the end of the protective tube at the will of the animal. The tentacles themselves may be expanded, giving a beautiful bell-shaped, flower-like appearance. They are hollow and are covered with fine hair-like cilia. They are muscular and can be bent and straightened at will. By their combined action currents in the water are set up toward the mouth, situated just beneath the lophophore. Minute organisms are thus swept in as food.

The Bryozoa increase by a process of budding which gives rise

to the branched stalks. There is also sexual reproduction. Statoblasts, or winter eggs, form within the body and escape after the death of the animal. They are sometimes formed in such abundance as to form patches of scum upon the surface of a pond. The various forms of these statoblasts assist in the classification of the Bryozoa.

Description of Common Forms. — The following are some of the important fresh-water genera. There are many marine forms.

Plumatella. — Zoary confervoid, brown-colored, branched, tubular; branches distinct. Lophophore crescent-shaped. Tentacles numerous, arranged in a double row. Statoblasts elliptical, with a cellular dark-brown annulus, but no spines. (Pl. XVIII, Fig. 6.)

Fredericella. — Zoary tubular, branched, brown-colored. Lophophore circular. Tentacles about 24, arranged in a single row. Statoblasts elliptical or subspherical, smooth, no spines, without a cellular annulus. (Pl. XVIII, Fig. 4.)

Paludicella. — Zoary tubular, diffusely branched, having the appearance of brown club-shaped cells joined end to end; apertures lateral, near the broad ends of the cells. Lophophore circular. Tentacles 16, arranged in a single row. Statoblasts elliptical, without spines, with a cellular bluish-purple annulus. (Pl. XVIII, Fig. 5.)

Pectinatella. — Zoary massive, gelatinous, fixed. Polypids protruding from orifices arranged irregularly upon the surface. Tentacles numerous. Statoblasts circular, with a single row of double hooks, not forked at the tips, as in *Cristatella*. Common. (Pl. XVIII, Fig. 7.)

Cristatella. — Zoary a mass of jelly, the polypids arranged on the outside, and the tentacles extended beyond the surface. The jelly mass is usually long and narrow and has the power of moving slowly, creeping over submerged objects. Tentacles numerous, pectinate upon two arms. Statoblasts circular, with two rows of double hooks having forked tips. Rare.

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CHAPTER XXX

PORIFERA

The fresh-water Porifera or sponges are not of sufficient importance in water supplies to warrant an extended description in this work. They differ materially from the marine sponges, which make up by far the greater part of the porifera.

Anatomy and Physiology. — The fresh-water sponge is an agglomeration of animal cells into a gelatinous mass, often referred to as the "sarcode." Embedded in the sarcode and supporting it are minute siliceous needles, or spicules. These skeleton spicules interlace and give the sponge mass a certain amount of rigidity. The sponge grows in flat patches upon the sides of water pipes and conduits and upon submerged objects in ponds and streams; or it extends outward in large masses or in finger-like processes that sometimes branch. Its color when exposed to the light is greenish or brownish, but in the dark places of a water supply system its color is much lighter and is sometimes creamy white. The sponge feeds upon the microscopic organisms in water, which are drawn in through an elaborate system of pores and canals. If these pores become choked up with silt and amorphous matter the organism dies. For this reason sponge patches are more abundant upon the top and sides of a conduit than upon the bottom.

At certain seasons the fresh-water sponges contain seed-like bodies known under the various names of gemmules, ovaria, statoblasts, statospheres, winter-buds, etc. They are nearly spherical and are about 0.5 mm. in diameter. They have a chitinous coat that encloses a compact mass of protoplasmic globules. In this coat there is a circular orifice, known as the foraminal aperture, through which the protoplasmic bodies make their exit at time of germination. In most species the chitinous coat is surrounded by a "crust" in which are embedded minute spicules, called the "gemmule spicules," to distinguish them from the "skeleton spicules," referred to above. There is a third kind of spicule known as the "dermal spicule" or the "flesh spicule." They lie upon the outer lining of the canals in the deeper portions of the sponge. They are smaller than the skeleton spicules and are not bound together. Dermal spicules are not found in all species.

The skeleton spicules differ somewhat in different species. They

have a length of about 250 μ . They are usually arcuate and pointed at the ends. They may be smooth or covered with spines (Pl. XVIII, Fig. 9). These skeleton spicules of sponge are commonly observed in the microscopical examination of surface waters. The gemmule spicules differ in character in different genera and species. Their characteristics are used, therefore, in classifying the fresh-water sponges.

Common Forms. — Potts has described a number of different genera of fresh-water porifera, among which are *Spongilla*, *Meyenia*, *Heteromeyenia*, *Tubella*, *Parmula*, *Carterius*, etc. The first two are the most important. They are sometimes given the rank of sub-families.

Spongilla is a green, branching sponge. The skeleton spicules are smooth and fasciculated. The dermal spicules are fusiform, pointed, and entirely spined. The gemmule spicules are cylindrical, more or less curved, and sparsely spined — the spines often recurved. (Pl. XVIII, Fig. 8.)

Meyenia is usually sessile and massive. The skeleton spicules are fusiform-acerate, abruptly pointed, coarsely spined except near the extremities; spines subconical, acute. The dermal spicules are generally absent. The gemmule spicules are irregular, birotulate, with rotules produced.

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CHAPTER XXXI

MISCELLANEOUS ORGANISMS

The importance of many of the higher aquatic organisms, both plants and animals, has been discussed in Chapter XII. Many of them have there been mentioned by name and some have been described. Apart from this phase of aquatic microscopy the miscellaneous higher animals and plants that one is likely to observe in a microscopical examination of drinking water are so varied, and they are of such little practical importance in the interpretation of a drinking water analysis, that their description here is not warranted. It is sufficient to mention the names of a few common forms.

Common Organisms. — Of the Vermes the following may be noted: *Anguillula*, a small, colorless thread-worm like the vinegar-eel (Pl. XIX, Fig. 1); *Gordius*, the common hair-snake; *Nais*, an annulate worm with bristles (Pl. XIX, Fig. 2); *Tubifex*, another bristle-bearing worm; *Chaetonotus*, an elongated worm-like organism with scales on its back (Pl. XIX, Fig. 3). Of the Arachnida: *Macrobiotus*, the water-bear (Pl. XIX, Fig. 4); and the *Acarina*, water-mites, or water-spiders (Pl. XIX, Fig. 5). Of the Hydrozoa: *Hydra*, a most interesting organism from a zoological standpoint (Pl. XIX, Fig. 6). Insect larvæ; *Corethra*, or the phantom larva; scales and fragments of insects; barbs of feathers; epithelium cells; ova of the Entozoa, Crustacea, Rotifera, etc.

Of the vegetable kingdom may be mentioned: fragments of *Sphagnum* or peat moss; *Myriophyllum*, or water milfoil; *Ceratophyllum*, or hornwort (Pl. XIX, Fig. 10); *Lemna*, or duck weed (Pl. XIX, Fig. 12); *Potamogeton*, or pond weed (Pl. XIX, Fig. 11); *Hippuris*, or mare's-tail; *Elodea*, or American water weed (Pl. XIX, Fig. 9); *Utricularia*, an insectivorous plant; pollen grains; plant hairs; fragments of vegetable fibers and tissue; fibers of cotton, hemp, etc.; starch grains, etc.

For the description of all these miscellaneous organisms and objects the reader is referred to more comprehensive books on zoology, botany, and general microscopy, and especially to "Fresh Water Biology," edited by Dr. Henry B. Ward and George C. Whipple.

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CHAPTER XXXII

ECOLOGICAL CLASSIFICATION OF MICROSCOPIC ORGANISMS

The preceding chapters of Part II have dealt chiefly with the structure of microscopic organisms and such other determinative characteristics as will aid in the identification of the forms of life commonly encountered in samples of water. Nothing has been said about the mutual relations between the different organisms and their environment, i.e. their ecology, a subject of great interest to sanitarians.

Kolkwitz and Marsson's Ecological System. — The sanitary ecology of aquatic organisms has been championed in particular by Kolkwitz and Marsson whose ecological system of saprobic organisms is made the basis of classification in this chapter. As explained in Chapter XII Kolkwitz and Marsson distinguished three broad zones of existence, called the polysaprobic zone, mesosaprobic zone, and oligosaprobic zone. These zones are characterized in the order of their statement by a gradual decrease in content of organic food stuffs and are variously described by their originators somewhat as follows:

Polysaprobic Zone. — This zone is characterized chemically by a wealth in high-molecular, decomposable, organic food matters (albumens and carbohydrates). These are derived from the waste waters that are discharged into lakes and streams by cities and industries. Disintegrating and splitting processes predominate, due to rapid disappearance of dissolved oxygen from the water. Black sludge deposits accumulate on the bottom.

From a biological standpoint this zone is marked by the occurrence of tremendous numbers of individuals, belonging, however, to only a few classes of organisms. Among them are particularly the schizomycetes (higher and lower bacteria) and the bacteriverous protozoa (colorless flagellates and ciliates). Fish are absent.

Mesosaprobic Zone. — For convenience this zone is divided into two parts, called α -mesosaprobic and β -mesosaprobic. The first adjoins the polysaprobic zone, the second extends towards the cleaner water. In the α portion of this zone natural purification processes remain more or less turbulent but, in contrast to activity in the polysaprobic zone, they are accompanied by oxidation phenomena in which a few chlorophyllaceous organisms may take part. Mineralization is approached in the β -mesosaprobic zone.

Life in the mesosaprobic zone is commonly tolerant of dilute or imperfectly purified sewage and its products of decomposition. Many bacteria are still present. Blue-green and grass-green algæ and diatoms reappear, also higher plants. Animal organisms are numerous and varied. Worms, protozoa and rotifers inhabit this zone. Tolerant higher animals feed on the bottom.

Oligosaprobic Zone. — This is a zone of cleaner water in which mineralization has been completed. The water is practically saturated with oxygen, sometimes even supersaturated. The organic nitrogen content is low, and the water is clear and transparent.

Bacteria are reduced in number. Chlorophyllaceous plants predominate. Protozoa and rotifera are joined by crustacea. Game fish make this zone their habitat. The plant and animal plankton of the cleaner inland lakes is oligosaprobic in character. The bottom deposits are usually clean but may partake of mesosaprobic qualities.

Not all organisms are definite in their ecological status. Some are found in more than one type of saprobic environment. Hence they are classified as indifferent. Organisms of this kind cannot serve as indicators of the conditions of existence obtaining in the water or in the mud deposits. A large group of organisms, furthermore, is not found at all where saprobic conditions occur. The flora and fauna of springs and pure mountain streams belong to this "katarobic" group and are so designated.

Key to Classification. — In order that the ecological status of various organisms may be determined quickly the following method of classification has been adopted in this chapter: Organisms are grouped in accordance with the scheme followed in the preceding chapters of Part II. Genera and species are arranged alphabetically. Their ecological character is indicated by the following letters:

- P = Polysaprobic
- M = Mesosaprobic (also α M and β M)
- O = Oligosaprobic
- K = Katarobic

Appended to these letters are subscript numbers which indicate the source of assignment of the organism. The numbers correspond to those used in listing the authorities.* For example: *Anabæna flos-aquæ* O_{1, 3, 15, 19} signifies that this organism is listed as oligosaprobic by Kolkwitz and Marsson, 1908; by Kolkwitz, 1911; by Hentschel, 1923; and by Schoenichen, 1925.

* See p. 556.

Whenever workers have not used the same ecological system as Kolkwitz and Marsson, the organisms described by them have been assigned to what appeared to be the class most closely conforming to the description of the environment in which they were encountered. Thus organisms found in sludge digestion tanks have been listed as polysaprobic, while organisms occurring on sprinkling filters have been designated mesosaprobic.

A study of the classification will show that there is as yet an appreciable lack in agreement as to the true status of many organisms. Relatively few species can be placed definitely in a particular environment: they are the indicator organisms, type organisms, or living reagents whose presence in a sample of water is a valuable diagnostic test of water conditions.

The ever-increasing interest in the conservation of our water resources should yield ecological information that will add greatly to the relatively meager or indefinite information now available. Many of the species listed in this classification are European forms. A North American ecology is much needed.

ALGÆ

Cyanophyceæ *Blue-green Algae*

- | | |
|--|--|
| Anabæna flos-aquæ, O ₁ , 3, 15, 19 | Oscillatoria antliaria, β M ₁ |
| spiroides, O ₁ , 19 | brevis, α M ₁ |
| Aphanothece luteola, P ₁₉ | chalybea, α M ₁ |
| microscopica, P ₁₉ | chlorina, α M ₁ , 3 |
| Aphanizomenon flos-aquæ, β M ₁ , 3, 15, 19 | formosa, α M ₁ , 6 |
| Arthrospira jenneri, P ₁ , 6, 19; α M ₁ , 6, 19 | lauterborni, P ₁₉ |
| Calothrix parietina, O ₁ , 19 | limosa, α M ₇ ; β M ₁ , 3, 6, 9 |
| Clathrocystis æruginosa, O ₁ , 9, 15 | princeps, α M ₁ , 19 |
| Celosphærium kützingianum, O ₁ | putrida, P ₁₉ ; α M ₁ |
| Dactylococcopsis acicularis, O ₁₉ | rubescens, O ₁ , 19 |
| raphidioides, O ₁ | splendida, α M ₁ , 19 |
| Glaucothrix gracillima, O ₁ | tenuis, P ₁₅ ; α M ₁ , 6, 19 |
| Gomphosphæria lacustris, O ₁ , 19 | Phormidium autumnale, α M ₁ , 6 |
| Merismopedia convoluta, O ₁ | faveolarum, α M ₁ |
| elegans, O ₃ | inundatum, O ₁ |
| glaucæ, O ₁ , 6 | papyraceum, O ₁ |
| Microcoleus subtorulosus, O ₁ | subfuscum, β M ₁ |
| Microcystis incerta, O ₁ | uncinatum, α M ₁ , 3; β M ₄ |
| Oscillatoria species, M ₁₄ | Polycystis æruginosa, O ₃ |
| agardhi, O ₁ | Spirulina jenneri, P ₃ ; M ₃ |
| angina, O ₁ | |

Chlorophyceæ Green Algæ

- Actinastrum hantzschii*, O₁, 16, 19
Ankistrodesmus polymorphus, β M₁;
 O₁, 3, 19
Brachionococcus chlorelloides, β M₁₉
Bulbochaete setigera, O₁, 3
Chaetophora elegans, O₁, 6, 19
Chara species (all species), O₃
 foetida, O₃
 fragilis, O₃
Chlorella infusumum, α M₁
 kolkwitzii, β M₁₉
 minor, β M₁₉
 oblonga, β M₁₉
Chlorococcum botrytis, β M₁, 19
Chlorogonium euchlorum, β M₁, 19
Chlorosphaera limicola, β M₁
Cladophora crispata, β M₁, 3, 6, 19
 glomerata, O₁, 4, 6, 9, 19
Closterium acerosum, β M₁, 3, 6, 19
 areolatum, O₁
 diana, O₁, 19
 chrenbergi, O₁, 3, 19
 leibleini, β M₁, 19
 lunula, O₁, 19
 moniliferum, β M₁₉
 peracerosum, β M₆
 parvulum, β M₁, 19
Cœlastrum microporum, O₁, 19
 reticulatum, O₁
Coleochaete pulvinata, O₁, 19
Cosmarium botrytis, β M₁
Dictyosphaerium ehrenbergianum, β M₁, 19
 pulchellum, β M₁, 16, 19
Dimorphococcus lunatus, O₁
Draparnaldia glomerata, O₁, 19
 plumosa, O₁, 19
Eudorina elegans, O₁, 3, 6, 9, 16, 19
Hydrodictyon utriculatum, O₁, 3, 10, 19;
 (β M₃ in masses)
Micrasterias rotata, O₃
Microthamnion kützingerianum, β M₁, 19
Mougeotia genuflexa, O₁, 6, 19
Nannochloris bacillaris, β M₁₉
 coccoides, β M₁₉
Edogonium species, β M₁, 6
Pandorina morum, O₁, 16, 19
Pediastrum boryanum, β M₁, 3, 9, 19; O₃
 duplex, O₁, 19
 kawraiskyi, O₁
 rotula, O₁
 tetras, O₁, 19
Prasiola crispa, α M₆
Protococcus species, M₁₄
 botryoides, O₁
Rhizoclonium hieroglyphicum, O₁, 6, 19
Richteriella botryoides, O₁, 6
Scenedesmus acuminatus, β M₁
 acutus, β M₃
 bijugatus, β M₁
 obliquus, β M₁
 quadricauda, β M₁, 6, 16
Schizomeris leibleini, M₇; O₇
Selenastrum bibrainum, β M₁, 19
Sphaerocystis schrœteri, O₁
Spirogyra crassa, β M₁, 3, 19
 gracilis, O₁, 19
 irregularis, O₁
 nitida, O₁, 19
 porticalis, β M₁, 19
Staurostrum tetracerum, O₁
Stichococcus bacillaris, α M₂, 6; β M₁, 6
Stigeoclonium species, M₁₄
 lubricum, β M₇
 tenuis, α M₁, 3, 6, 9; β M₁, 3, 7
Tetraspora explanata, O₁
 gelatinosa, O₁, 6
Tribonema bombycina, β M₁, 3, 6
Ulothrix subtilis, α M₁, 6; β M₁, 6; O₁, 6
 variabilis, O₁, 6
 zonata, β M₃; O₁, 3, 7
Vaucheria species, O₁, 6
 sessilis, β M₁, 6, 19

Xanthophyceæ Yellow-green Algæ

- Botryococcus brauni*, O₁, 6

Diatomaceæ Diatoms

- Achnantes exilis*, O_1 , 19
Amphora ovalis, O_1 , 19
Asterionella formosa, O_1 , 3, 6, 9, 10, 19
Bacillaria paradoxa, O_1 , 6, 19
Caloneis amphibæna, βM_{19}
Cocconeis pediculus, βM_3 ; O_3
Cyclotella comta, O_1
 kützingiana, O_1
 meneghiniana, O_1 , 6
Cymatopleura elliptica, O_1 , 19
 solea, O_1 , 3, 19
Cymbella cistula, O_1 , 6, 19
 ehrenbergi, O_1
 lanccolata, O_1 , 19
 prostrata, O_{19}
 ventricosa, O_{19}
Diatoma vulgare, βM_1 , 6, 16
Encyonema prostratum, O_1
 ventricosum, O_1
Epithemia sorex, O_1
 turgida, O_1 , 19
 zebra, O_1 , 19
Eunotia arcus, O_1 , 6
Fragillaria construens, O_1 , 19
 crotonensis, O_9
 mutabilis, O_1 , 19
 virescens, O_1 , 6, 19
Gomphonema acuminatum, O_1 , 3, 6, 19
 angustatum, O_1 , 19
 capitatum, O_1
 constrictum, O_1 , 19
 olivaceum, βM_1 , 3, 6, 9, 19; O_9
 parvulum, βM_1 , 6, 19
 tenellum, βM_1
Hantzschia amphioxys, αM_1 , 3, 6
Melosira ambigua, O_1
 arenaria, O_1 , 3, 19
 binderiana, O_1 , 3
 crenulata, O_1
 granulata, βM_7 ; O_1
 italica, O_1
 varians, αM_3 ; βM_1 , 3, 6, 9
Meridion circulare, O_1 , 6, 19
 constrictum, O_{19}
Microneis minutissima, βM_1
Navicula ambigua, βM_1 , 6
 amphibæna, βM_1 , 6
 atomus, βM_1 , 19
Navicula brebissoni, βM_1 , 6
 clausi, O_1
 cryptocephala, βM_1 , 6, 9, 19
 cuspidata, βM_1 , 3, 6, 19
 dicephala, O_1 , 19
 gastrum, O_1 , 19
 gibba, O_1
 hungaria, O_1 , 19
 inflata, O_1
 iridis, O_1
 limosa, O_1 , 6
 major, O_1 , 6
 mesolepta, βM_1 , 6; O_1 , 6
 perpusilla, O_1
 radiata, O_1
 radiosa, βM_1 , 19
 rhynchocephala, βM_1 , 19
 viridis, O_1
 viridula, O_1 , 19
Neidium iridis, O_{19}
Nitzschia acicularis, βM_1 , 19
 amphioxys, αM_{19}
 communis, βM_1 , 6, 19
 dissipata, βM_1 , 19
 linearis, O_1 , 19
 palea, αM_1 , 6
 parvula, βM_1 , 19
 sigmoidea, O_1 , 19
 stagnorum, βM_1
 vermicularis, O_1
 vitrea, O_1
Pinnularia brebissoni, βM_{19}
 gibba, O_{19}
 major, O_{19}
 mesolepta, βM_{19}
 viridis, O_3
Pleurosigma attenuatum, O_1 , 3, 19
Pleurostauron acutum, αM_{19}
Rhoicosphenia curvata, βM_1 , 6
Rhopalodia gibba, O_1
Stauroneis acuta, αM_1 , 6
 phœnicenteron, βM_1
Stephanodiscus hantzschianus, βM_1 , 6
Surirella biseriata, O_1 , 19
 ovalis, βM_1 , 6
 splendida, O_1 , 3, 6
Synedra actinastroides, βM_1 , 15
 acus, O_1 , 9

Synedra radians, βM_1 , 19
splendens, αM_1
ulna, O_1 , 19

Synedra vaucheriae, βM_1
Tabellaria fenestrata, O_1 , 2
flocculosa, O_1 , 3, 4, 6, 19

Rhodophyceæ Red Algae

Batrachospermum moniliforme, O_1 , 3, 19
Chantransia chalybea, O_3

Lemanea annulata, O_3
torulosa, O_1 , 3, 6, 19

FUNGI

Schizomycetes Fission Fungi

(Excluding the Bacteria)

Beggiatoa alba, P_1 , 3, 6, 9, 15, 19; M_{14}
arachnoidea, P_1 , 3, 19
leptomitiformis, P_1 , 19
Chromatium minutissimum, P_2 ; M_1
okeni, P_2 , 3, 6, 15; αM_6 , 15; M_2 , 3
vinosum, P_1 ; M_1
Clonothrix fusca, O_1 , 3, 15, 19
tenuis, P_{19}
Crenothrix polyspora, O_1 , 3, 6, 9, 19
Didymohelix ferruginea, O_1 , 3, 6, 9

Lamprocystis roseopersicana, P_2 , 3, 6;
 αM_6 ; M_2
Lampropedia hyalina, βM_1
Leptothrix ochracea, O_1 , 3, 6
Sphaerotilus dichotomus, αM_{15} ;
 βM_1 , 3, 6, 9, 19
natans, P_1 , 3, 6, 9, 10, 15, 19; αM_6 , 9, 19; M_1
roseus, P_1 , 3; M_1
Thiothrix species, P_{10}
nivea, P_1 ; αM_1 , 3, 6, 15, 19

Phycomycetes Molds

Endoblastoderma salmonicolor, αM_1
Fusarium species, αM_3
aqueductuum, αM_1 , 9
aurantiacum, αM_6
Leptomitius lacteus, P_{10} , 15; αM_1 , 3, 6, 9

Mucor species, P_3 ; αM_2 , 9; M_2
Oidium species, M_{14}
Penicillium species, M_{14}
Pythium species, M_{14}
Saprolegnia species, P_{10}

PROTOZOA

Sarcodina

Acanthocystis turfacea, βM_2 ; O_2
Actinophrys sol, αM_2 ; βM_2 , 6, 15; M_{20}
Actinosphaerium eichhorni, αM_2 ;
 βM_2 , 3, 15; M_{20}
Amoeba brachiata, βM_2
guttula, P_2 ; M_{18} , 16
limax, P_2 , 3, 15; M_3 , 13, 16
proteus, P_{17} ; M_{13} , 17; O_2 , 9
radiosa, βM_2 , 3; M_{16} , 20
verrucosa, βM_2 ; M_{20}
Arcella species, M_{13}
dentata, M_{20}
vulgaris, αM_2 ; βM_2 , 3, 6, 9; M_{20}

Centropyxis aculeata, βM_2 , 6; M_{20}
Chlamydothrys stercorea, P_{17} ; M_{20}
Clathrulina elegans, αM_2 ; βM_2
Cochliopodium bilimbosum, βM_2 ; M_{20}
pellucidum, αM_2 ; βM_2
Cryptodiffugia oviformis, αM_2
Cyphidium aurelium, O_2
Cyphoderia ampulla, O_2
Diffugia species, M_{18} ; O_2
acuminata, O_2 , 3
corona, βM_2 ; O_2
globulosa, βM_2 ; O_2
hydrostatica, O_2

Diffugia limnetica, O₂
 pyriformis, βM_2 ; M₂₀; O₂, 5
 urceolata, O₂

Dimastigamœba gruberi, P₁₇

Diplophrys archeri, αM_2

Euglypha species, M₁₃
 alveolata, P₁₇; βM_2 ; M₂₀
 globosa, O₂

Hartmannella hyalina, P₁₇; M₂₀

Lesquereusia spiralis, O₂

Microgromia socialis, βM_2 ; O₂

Pamphagus armatus, αM_2
 hyalinus, αM_2

Pamphagus mutabilis, βM_2 ; M₂₀

Pelomyxa palustris, αM_2 ; βM_2 , 15

Platoom stercoræum, βM_2

Protamœba primitiva, M₂₀

Raphidiophrys pallida, βM_2 ; O₂, 9

Sphærastrum fockei, βM_2

Trinema species, M₁₃

enchelys, αM_2

lineare, P₁₇; M₁₆

Vahlkampfia albida, P₁₇; M₂₀

guttula, P₁₇; M₂₀

limax, P₁₇; M₂₀

Mastigophora

Amphimonas cyclosum, O₆; K₆

fusiformis, αM_2 ; M₆

globosa, αM_2 ; M₆, 19; O₆, 19

Ancyromonas contorta, O₆

Anisonema acinus, βM_2 ; M₆; K₆

ovale, O₆; K₆

truncatum, O₆; K₆

variabile, O₆; K₆

Anthophysa steini, M₆

vegetans, P₁₇; αM_2 , 3, 4, 6, 9, 16; M₆

Astasia species, M₁₆

curvata, P₆, 19; M₆, 19

dangeardi, M₆, 19

distorta, αM_2

inflata, M₆, 19

klebsi, P₆, 19; M₆, 19

margaritifera, αM_2

ocellata, O₆, 19

Bikosœca dinobryoidea, O₆, 19; K₆

lacustris, βM_2 ; O₂, 5, 19; K₆

oculata, βM_2 ; O₂, 5, 19; K₆

Bodo alexeieffi, M₆

angustus, P₁₇; M₆, 19

caudatus, P₁₇; αM_2 ; M₆, 19

celer, βM_2 ; M₆, 19

compressus, M₆, 19

cruzi, P₆, 19

edax, P₁₇; M₆, 19

fusiformis, M₆

globosus, αM_2 , 3, 9; M₆, 19

lens, M₆, 19

ludibundus, M₆, 19

minimus, αM_2 ; M₆, 19

mutabilis, αM_2 ; M₆, 19

Bodo obovatus, M₆, 19

ovatus, P₁₇; αM_2 ; M₆, 19

putrinus, P₂, 6; M₆, 19

repens, βM_2 ; M₆, 19

rostratus, βM_2 ; M₆, 19

saltans, P₃, 4, 5, 19; αM_2 ; M₆, 5, 19

uncinatus, βM_2 ; M₆, 19

variabilis, M₆, 19

Bodopsis alternans, O₆; K₆

Carteria cordiformis, βM_1 , 19

obtusa, O₁

Ceratium hirundinella, O₁, 3, 6, 9

tetracerum, βM_1

Cercobodo agilis, P₆, 19; M₆, 19

alexieffi, M₆, 19

bodo, O₆, 19; K₆

caudatus, P₁₇

crassicauda, P₆, 17, 19; M₆, 19

digitalis, O₆, 19; K₆

laciniatgerens, M₆, 19

longicauda, P₂, 5, 17, 19; αM_2 ; M₆, 19

ovatus, P₁₇; O₆, 19

radiatus, P₆; αM_2

simplex, P₆

Cercomastix parva, P₆

Cercomonas clavata, αM_2

crassicauda, αM_2

Chilomonas paramecium, αM_2 , 6; βM_2

Chlamydomonas species, P₁₇

angulosa, O₁

debaryana, αM_1

brauni, βM_1 , 19

ehrenbergi, βM_1 , 19

intermedia, O₁, 19

- Chlamydomonas kuteinikowi*, βM_1
 longistigma, O_1
 pisciformis, O_1
 reinhardi, βM_1 , 3, 19
 reticulata, βM_1
 variabilis, O_1
Chloromonas reticulata, βM_{19}
Chromulina rosanoffi, O_1 , 3
Chrysococcus porifer, βM_{19}
Chrysosphaerella longispina, βM_1 , 19
Ciliophrys infusionum, αM_2
Clautriavia parva, P_{17}
Colacium vesiculosum, βM_1 , 19
Colloidietyum triciliatum, M_5 ; K_5
Cryptoglena chrenbergi, αM_{19} ; M_5 ; K_5
 pigra, αM_1
Cryptomonas species, βM_{15}
 erosa, βM_1 , 3
 mordstedti, αM_1
 ovata, βM_1
Dallingeria drysdali, αM_2 ; βM_2 ; M_5 , 19
Dimorpha alternans, βM_2 ; O_2
Dinema griseolum, O_5 ; I_{15}
Dinobryon species, O_1 , 6, 15
 sertularia, βM_9 ; O_3 , 9
Dinomonas tuberculata, M_5
 vorax, P_{17} ; M_5
Diplosiga frequentissima, βM_2 ; O_2
Distigma species, M_{16}
 proteus, P_{17} ; M_5 ; K_5
Entosiphon obliquum, M_5 ; K_5
 ovatum, M_5
 sulcatum, P_{17} ; βM_2 ; M_5 , 16; K_5
Euglena species, M_{16}
 acus, βM_1 , 6; M_5 , 19; O_5 , 19
 deses, βM_1 , 6, 19; M_5
 ehrenbergi, O_5 , 19; K_5
 fusca, O_5 , 19; K_5
 geniculata, O_1 , 5, 19; K_5
 gracilis, P_{17} ; M_{16}
 granulata, M_5 , 19; O_5 , 19
 haematodes, O_5 ; K_5
 intermedia, P_{17} ; M_5 , 16, 19
 minima, O_1 , 5; K_5
 mutabilis, P_{17} ; M_{16}
 oblonga, O_1 , 4, 19; K_5
 olivacea, M_5 , 19
 oxyuris, βM_1 , 19; M_5 ; K_5
 pisciformis, βM_1 ; M_5 , 19; O_5 , 19
 polymorpha, P_1 ; M_5 ; O_5
 quartana, βM_1 ; M_5 , 19
 sanguinea, O_5 , 19; K_5
 sociabilis, M_5
 spirogyra, βM_1 , 6; M_5 , 19; O_5 , 19
 tripteris, βM_1 , 19; M_5 ; K_5
 variabilis, O_5 , 19; K_5
 velata, βM_1 , 19; M_5
 viridis, P_1 , 3, 5, 6, 9, 16; αM_1 , 6, 9; M_5
Euglenopsis vorax, P_5 , 19; αM_2 ; M_5 , 19
Eutreptia viridis, M_5 , 19; K_5
Furcilla lobosa, M_5
Glenodinium gymnodinium, M_{19} ; O_{19}
Gonium sociale, βM_1
 tetras, βM_{19}
Gonyaulax apiculata, O_1
Gymnodinium palustre, M_{19} ; O_1 , 3, 19
Heteronema species, P_{17} ; M_{16}
 acus, M_5 , 19
 globiferum, O_5 ; K_5
 nebulosum, O_5 ; K_5
 spirale, O_5 ; K_5
 tremulum, αM_2 ; M_5 ; K_5
Hexamitus crassus, P_2 , 5, 19
 fissus, P_2 , 5, 19; αM_2 ; M_5 , 19
 fusiformis, P_2 , 5, 19; αM_2 ; M_5 , 19
 inflatus, P_2 , 3, 5, 17, 19; αM_2 ; M_5 , 5, 19
 pusillus, P_2 , 5, 19; αM_2 ; M_5
Lagenœca cuspidata, M_5 ; K_5
 obovata, O_5 ; K_5
Lepocinclis ovum, αM_1 ; O_5 , 19; K_5
 texta, αM_1 ; M_5 ; O_5
Mallomonas acaroides, O_1
 producta, O_1
Mastigamoeba species, M_{16}
 aspera, βM_2 ; O_5 , 19; K_5
 bütschlii, O_5 , 19; K_5
 invertens, βM_2 ; O_5 , 19; K_5
 limax, P_5 ; βM_2
 longifilum, P_{17} ; K_5
 polyvacuolata, βM_2
 ramulosa, O_5 , 19; K_5
 reptans, P_{17} ; K_5
 trichophora, P_5 , 19
Mastigella commutans, O_5 , 19; K_5
 radicula, P_5 ; M_5
 simplex, P_{17} ; M_5 ; K_5
Menoidium falcatum, βM_2
 incurvum, P_{17} ; M_5 , 19
 pellucidum, αM_2 ; M_5 , 19
 tortuosum, M_5

- Monas amoebina*, P₁₇
arhabdomonas, αM_2 ; M₅, 19
dangeardi, M₅
minima, P₁₇; M₅, 19
socialis, M₅, 19
vivipara, αM_2 , 6, 7; M₅, 19
vulgaris, αM_2 , 6
Multicilia lacustris, O₅; K₅
palustris, O₅; K₅
Notosolenus species, M₁₆
orbicularis, P₁₇
Oicomonas mutabilis, P₂, 5, 19
quadrata, M₅, 19
rostrata, M₅, 19
socialis, P₅, 17; βM_2
steini, P₅, 19
termo, P₅, 7, 17, 19; αM_2 , 6, 7; M₅, 19
Peranema species, M₁₆
granuliferum, O₅; K₅
trichophorum, P₁₇; αM_2 , 3, 6; M₅, 16; K₅
Peridinium berolinense, O₁
bipes, O₁
cinctum, O₁, 6
minimum, O₁
quadridentis, O₁
tabulatum, O₁
Petalomonas abscissa, O₅; K₅
angusta, O₅; K₅
carinata, P₁₇; K₅
inflexa, O₅; K₅
mediocanellata, P₁₇; M₅; K₅
sexlobata, O₅; K₅
steini, O₅; K₅
Phacotus lenticularis, O₁₉
Phacus species, O₁₆
alata, O₅, 19; K₅
brevicaudata, O₅, 19; K₅
caudata, βM_1 , 5, 19; K₅
hispidula, O₅; K₅
longicauda; O₁, 5, 19; K₅
parvula, O₁
pleuronectes, O₁, 3, 5, 19; K₅
pyrum, O₁, 5, 19; K₅
triquetra, O₅; K₅
Phalansterium consociatum, O₅; K₅
digitatum, O₅; K₅
Phialonema cyclostomum, βM_1
Phyllomitus amylophagus, αM_2 ; M₅
Phyllomitus undulans, M₅
Phyllomonas contorta, O₁₉
Platytheca microspora, P₁₇; O₅
Pleuromonas jaculans, P₁₇; αM_2 ; M₅, 19
Polytoma uvella, P₁, 3, 17, 19
Pteridomonas scherffeli, O₅; K₅
Pteromonas protracta, O₁₉
Rhynchomonas nasuta, αM_2 ; M₅, 19
Salpingoeca bütschlii, M₅
frequentissima, O₅; K₅
fusiformis, O₅; K₅
marssoni, P₁₇; K₅
vaginicola, P₅
Seytomonas major, P₅
pusilla, P₅, 19; αM_2
Spharæcea volvox, βM_2 ; K₅
Sphenomonas quadrangularis, M₅; K₅
teres, M₅; K₅
Spondylomorom quaternarium, αM_1 , 19
Spongomonas intestinum, αM_2 ; βM_2 ;
M₅, 19; O₅, 19
sacculus, O₅; K₅
uvella, O₅; K₅
Sterromonas formicina, M₅, 19
Synura uvella, αM_5 ; βM_1 , 10, 19; M₅;
O₅, 9, 15
Tetramitus descissus, αM_2 ; P₁₇; M₁₉
pyriformis, αM_2 ; M₅, 19
rostratus, αM_2 ; M₅, 19
sulcatus, αM_2 ; M₅, 19
Trachelomonas species, βM_{15}
euchlora, M₅; K₅
hispidula, βM_1 ; M₅; K₅
reticulata, P₅
rugulosa, M₅; K₅
volvocina, βM_1 ; M₅; K₅
Trepomonas agilis, P₅, 17, 19; αM_2 ; βM_2 ;
M₅, 19
rotans, P₂, 5, 19; αM_2 ; M₅, 19
steini, P₅, 19; αM_2
Trigonomonas compressa, αM_2 ; M₅, 19
Tropidoscyphus octocostatus, O₅; K₅
Urcolus cyclostomus, M₅; K₅
Urophagus angustus, M₅
rostratus, αM_2 ; M₅
Uroglena volvox, O₁, 19
Volvox globator, O₁, 3, 6, 19

Infusoria

- Acineta grandis*, βM_2 , 15
Amphileptus carchesii, αM_2
 claparedi, P_2 , 3; αM_2 , 3
Amphisia species, M_{16}
Aspidisca species, M_{13} , 16
 costata, P_{17} ; αM_2 ; βM_2
 lynceus, αM_2 ; βM_2
 turrita, βM_{16}
Astylozoön fallax, βM_2
Balantiophorus minutus, βM_2 ; O_2
Blepharisma species, M_{13} , 16
 lateritium, βM_2
Bursaria species, M_{13}
 truncatella, βM_2
 tentaculata, βM_2
Cænomorpha species, M_{13}
 medusula, αM_2
Carchesium species, M_{16}
 epistylis, βM_2 , 6
 lachmanni, P_7 ; αM_2 , 3, 6, 9
 polypinum, αM_{15} ; βM_2 , 16; O_2 , 6
Chaënia species, M_{13}
Chasmatostoma reniforme, βM_2
Chilodon species, P_{17} ; M_{13} , 16
 cucullus, αM_2 , 15; βM_2 , 3, 15
 uncinatus, αM_2 ; βM_2
Cinetochilum margaritaceum, P_{17} ; βM_2
Climacostomum virens, βM_2
Codonella lacustris, βM_2 ; O_2
Coleps hirtus, αM_2 , 3; βM_2 , 3, 6, 9, 15
Colpidium species, P_{17} ; M_{16}
 colpoda, P_3 ; αM_2 , 3, 6
Colpoda species, M_{13} , 16
 cucullus, P_{17} ; αM_2 , 15
 inflata, P_{17}
 parvifrons, αM_2 ; βM_2
 steini, αM_2 ; βM_2
Condyllostoma vorticella, βM_2 ; O_2
Cothurnia crystallina, βM_2
Cyclidium species, M_{13} , 16
 glaucoma, P_{17} ; αM_2
Cyclogramma rubens, αM_2
Didinium nasutum, βM_2 , 15
Dileptus trachelioides, O_2
Disematostoma bütschlii, βM_2
Dysteropsis minuta, βM_2
Enchelys species, M_{13}
 pupa, βM_2
 Enchelys silesiaca, βM_2
Epistylis galca, αM_2
 coarctata, αM_2 , 6
 plicatilis, P_7 ; αM_2 ; βM_2
 umbellaria, βM_2
Euplotes species, M_{13} , 16
 charon, αM_2 ; βM_2
 patella, αM_2 ; βM_2 , 19
Frontonia species, M_{13} , 16
 acuminata, βM_2 ; O_2
Gastrostyla mystacea, αM_2
Gerda gians, αM_2
Glaucoma species, M_{13} , 16
 scintillans, P_{17} ; αM_2 , 3, 9, 15; βM_2
Halteria species, M_{13} , 17
 grandinella, βM_2 , 3, 15
Hexotricha caudatum, P_{17}
Holophrya species, P_{17} ; M_{13} , 16
 ovum, βM_2 ; O_2
Lacrymaria species, M_{13} , 16
 olor, O_2
Lagynus elegans, βM_2
Lembadion bullinum, βM_2
Lembus species, M_{16}
Leucophrydium putrinum, αM_2
Lionotopsis species, M_{13} , 16
Lionotus species, M_{13} , 16
 varsaviensis, αM_2
Loxocephalus granulatus, αM_2
Loxodes species, M_{16}
 rostrum, βM_2
Loxophyllum species, M_{16}
 armatum, βM_2
 faciola, P_{17} ; αM_2 ; βM_2
 lamella, βM_2
 meleagris, αM_2 ; βM_2
Metopus species, M_{13} , 16
 contortus, βM_2
 pyriformis, βM_2
 sigmoides, P_{17} ; αM_2 ; βM_2
Microthorax species, M_{16}
Nassula species, M_{16}
 elegans, βM_2 , 6
 ornata, βM_2
Opercularia species, M_{13} , 16
 berberina, P_{17}
Ophrydium versatile, O_2 , 3
Ophryoglena atra, O_2

- Opisthodon niemeccensis*, βM_2
Oxytricha species, P_{17} ; $M_{13, 16}$
 fallax, αM_2
 ferruginea, O_2
 pellionella, $\alpha M_2, 15$
Paramecium species, $M_{13, 16}$
 aurelia, $\alpha M_2, 15$; $\beta M_2, 15$
 bursaria, $\alpha M_2, 6$
 caudatum, $P_3, 17$; $\alpha M_2, 6, 9, 15$; M_3
 putrinum, $P_2, 3, 6, 7, 17$; M_2
Plagiopyla nasuta, P_{17} ; βM_2
Pleuronema species, M_{16}
 chrysalis, P_{17} ; βM_2
Podophrya species, βM_3 ; M_{16}
 carchesii, βM_2
 fixa, αM_2 ; βM_2
 quadripartita, βM_2
Prorodon species, P_{17} ; M_{16}
 faretus, βM_2
 platyodon, βM_2
Rhabdostyla ovum, O_2
Saprodinium putrinum, P_{17}
Saprophylus species, M_{13}
Spathidium species, M_{13}
Sphaerophrya species, βM_2
 pusilla, βM_2
Spirostomum species, $M_{13, 16}$
 ambiguum, $\alpha M_2, 8, 15$; $\beta M_2, 19$
 teres, βM_2
Staurophrya elegans, O_2
Stentor species, $M_{13, 16}$
 coeruleus, $\alpha M_2, 3, 6, 15$
 igneus, βM_2
 niger, βM_2
 polymorphus, αM_3 ; $\beta M_2, 3, 6, 9, 15$; O_2
 reseli, αM_2 ; $\beta M_2, 3, 15$
Stichotricha species, M_{13}
Strombidium adherens, O_2
 turbo, βM_2 ; O_2
Stylonychia species, $M_{13, 16}$
 histrio, O_2
 mytilus, $\alpha M_2, 15$; $\beta M_2, 15$
 pustulata, βM_2 ; O_2
Suctorina genera, M_2 (mostly)
Thylakidium truncatum, βM_2
Tintinnidium fluviatile, $\beta M_2, 15$; O_2
Trachelius elephantinus, O_2
 ovum, βM_{15}
Trachelophyllum lamella, βM_2
 pusillum, βM_2
Trimyema compressa, P_{17}
Trochilia palustris, αM_2 ; βM_2
Urocentrum turbo, βM_2
Uroleptus species, βM_{15} ; M_{16}
 musculus, αM_2 ; βM_2
 piscis, αM_2 ; βM_2
Uronema species, M_{16}
 griseolum, βM_2
 marinum, βM_2
Urostyla weissi, P_2 ; αM_2
Urotricha furcata, $\alpha M_2, 6$
 langenula, βM_2
Vorticella species, P_{17} ; $M_{13, 16}$
 campanula, $\beta M_2, 15$
 citrina, $\beta M_2, 15$
 convallaria, $\alpha M_2, 3$
 microstoma, $P_2, 3, 6, 7, 9, 15$; M_3
 nebulifera, $O_1, 3$
 patellina, βM_2
 putrina, $P_2, 6$
Zoöthamnium arbuscula, βM_2

ROTIFERA

- Anapus ovalis*, βM_2 ; O_2
 testudo, βM_2 ; O_2
Anuraea aculeata, $\beta M_2, 3, 6, 9, 15$; $O_2, 3$
 cochlearis, $\beta M_2, 3, 15$; $O_2, 3$
 hypelasma, O_2
 tecta, βM_{15}
Asplanchna species, βM_3 ; O_3
 brightwelli, $O_3, 3$
 priodonta, $\beta M_3, 3$; $O_3, 3$
Atrochus tentaculatus, βM_3
Brachionus angularis, αM_2 ; βM_2
 bakeri, βM_2
 pala, $\beta M_2, 9, 15$
 militaris, αM_3 ; βM_2
 rubens, βM_2
 urceolaris, βM_2
Callidina elegans, $\alpha M_3, 3, 6$
Cathypna luna, βM_2
Colurus bicuspidatus, αM_2 ; βM_2
 deflexus, βM_2

Conochilus unicornis, βM_2
Diaschiza semiaperta, αM_2 ; βM_2
 tenuior, βM_2
Diglena biraphis, αM_2 ; βM_2
 catellina, αM_2 ; βM_2
 caudata, αM_2 ; βM_2
 forcipata, αM_2 ; βM_2
Dinocharis pocillum, βM_2 ; O_2
 tetractis, βM_2 ; O_2
Diplax compressa, αM_2
 trigona, αM_2
Diplois daviesiae, αM_2
Diurella stylata, O_2
Euchlanis dilatata, βM_2 ; O_2
 triquetra, βM_2
Floscularia atrochoides, βM_2
 cornuta, O_2
Furcularia forficula, βM_2
 gibba, βM_2
 gracilis, αM_2 ; βM_2
 reinhardtii, βM_2
Gastropus styliifer, βM_2 ; O_2
Gastroschiza flexilis, βM_2 ; O_2
Hydatina senta, αM_2 , 3; βM_2
Melicerta ringens, βM_2 , 6
Metopidia oxysternum, βM_2
 lepadella, βM_2
Monostyla cornuta, βM_2
 lunaris, βM_2
Noteus quadricornis, βM_2
Notholca acuminata, βM_2
 foliacea, βM_2
 labis, βM_2

Notholca longispina, O_2 , 3, 6
 scapha, O_2
 striata, βM_2
Pedalion mirum, O_2
Philodina erythrophthalma, βM_2
 mcgalotrocha, βM_2
 roseola, αM_2 ; βM_2
Ploesoma truncatum, βM_2 ; O_2
Polyarthra species, βM_2 ; O_2
 platyptera, βM_2 , 9; O_2
Pompholyx complanata, O_2
 sulcata, βM_2
Proales tigridia, βM_2
Pterodina patina, αM_2 ; βM_2
Rattulus capucinus, βM_2 ; O_2
Rotifer actinurus, P_2 , 3, 7, 16; αM_2 ; M_2
 macrurus, βM_2
 tardus, βM_2
 vulgaris, αM_2 , 3, 6, 9, 16; βM_2 , 3, 6, 9, 11, 18
Salpina brevispina, O_2
 macrocantha, βM_2
 mucronata, αM_2 ; βM_2
Scaridium longicaudum, βM_2
Schizocerca diversicornis, βM_2 ; O_2
Stephanops unisetatus, βM_2
Synchata species, βM_2 ; O_2
 pectinata, βM_2 , 9; O_2
 tremula, βM_2 ; O_2
Taphrocampa selenura, βM_2
Triarthra breviseta, O_2
 longiseta, αM_2 , 3, 16; βM_2 , 3
 mystacina, βM_2

CRUSTACEA

Cladocera Water Fleas

Acroperus harpæ, O_2
Alona costata, O_2
 guttata, βM_2 ; O_2
Bosmina coregoni, O_2 , 3, 9
 longirostris, βM_2 , 16; O_2
Ceriodaphnia reticulata, βM_2 ; O_2
Chydorus sphaericus, βM_2 , 15
Daphnia cucullata, O_2
 hyalina, O_2
 longispina, αM_2 ; βM_2 , 9, 16; O_2
 magna, αM_2 ; βM_2

Daphnia pulex, αM_2 , 3; βM_2 , 3, 6
 schæfferi, αM_2 ; βM_2
Diaphanosoma brachyurum, O_2
 leuchtenbergianum, O_2
Holopedium gibberum, O_2
Leptodora kindti, O_2 , 3, 15
Moina rectirostris, βM_2
Pleuroxus excisus, βM_2
Sida cristallina, O_2
Simocephalus vetulus, O_2

Copepoda

<i>Canthocamptus minutus</i> , O ₂	<i>Cyclops serrulatus</i> , βM_2 ; O ₂
<i>staphylinus</i> , βM_2 , 3	<i>strenuus</i> , αM_2 , 3; βM_2 , 3
<i>Cyclops albidus</i> , O ₂	<i>viridis</i> , O ₂ , 4
<i>bicuspidatus</i> , O ₂	<i>Diaptomus castor</i> , βM_2
<i>brevicornis</i> , αM_2 ; βM_2	<i>gracilis</i> , O ₂
<i>fimbriatus</i> , βM_2	<i>graciloides</i> , O ₂ , 3
<i>fuscus</i> , O ₂	<i>laciniatus</i> , O ₂
<i>leuckarti</i> , βM_2	<i>Eurytemora velox</i> , O ₂
<i>oithonoides</i> , O ₂	<i>Nauplius</i> of <i>Cyclops strenuus</i> , αM_2 ;
<i>phaleratus</i> , βM_2	βM_2 ; O ₉

Ostracoda

<i>Candona candida</i> , βM_2	<i>Cypris</i> species, βM_2 ; O ₂
<i>Cypria ophthalmica</i> , βM_2	<i>incongruens</i> , βM_2 ; O ₂
<i>Cypridopsis vidua</i> , βM_2	<i>virens</i> , βM_2 ; O ₂

Malacostraca Higher Crustaceans

<i>Asellus aquaticus</i> , αM_2 , 3, 6, 9, 13;	<i>Gammarus fluviatilis</i> , βM_2 , 3, 6, 13
βM_2 , 3, 6, 9, 15	<i>pulex</i> , O ₂ , 3, 6, 9
<i>communis</i> , Indifferent ₁₀	<i>Hyalella</i> species, O ₁₀
<i>Astacus fluviatilis</i> , O ₂	<i>knickerbockeri</i> , βM_7
<i>Cambarus immnis</i> , Indifferent ₁₀	<i>Niphargus puteanus</i> , O ₂

Bryozoa Moss Animalcules

<i>Cristatella mucedo</i> , O ₂	<i>Plumatella</i> , <i>fungosa</i> , αM_{15} ; βM_2 , 15
<i>Fredericella sultana</i> , O ₂	<i>repens</i> , βM_2 , 3, 6
<i>Paludicella ehrenbergi</i> , O ₂	

Porifera Sponges

<i>Ephydatia fluviatilis</i> , αM_{15} ; βM_2 , 6, 15	<i>Spongilla fragilis</i> , βM_2
<i>muelleri</i> , βM_2	(Most sponges are indifferent)
<i>Euspongilla lacustris</i> , βM_2 , 3, 6, 9	

MISCELLANEOUS ORGANISMS**Archigoniatae Higher Plants**

<i>Amblystegium riparium</i> , O ₁ , 9	<i>Lemna polyrrhiza</i> , βM_1 , 3
<i>Ceratophyllum demersum</i> , βM_1 , 6	<i>trislula</i> , O ₂
<i>Elodea canadensis</i> , βM_1 , 3, 6, 9	<i>Nuphar luteum</i> , βM_3 ; O ₁ , 3
<i>Fontinalis antipyretica</i> , O ₁ , 3, 6	<i>Nymphaea alba</i> , βM_3 ; O ₁ , 3
<i>Hildenbrandia rivularis</i> , βM_1	<i>Phragmites communis</i> , αM_3 ; βM_3 ; O ₉
<i>Hypnum riparium</i> , O ₃	<i>Potamogeton crispus</i> , βM_3 ; O ₁ , 3, 6
<i>Isoetes lacustris</i> , O ₁	<i>pectinatus</i> , O ₁ , 6
<i>echinosporum</i> , O ₁	<i>perfoliatus</i> , βM_1 ; O ₂
<i>Lemna minor</i> , βM_1 , 6	<i>Salvinia natans</i> , O ₁

Hydrozoa

Cordylophora lacustris, O₂, 16
Hydra species, βM_{16} ; O₁₆
oligactis, βM_3 ; O₂, 3, 6

Hydra polypus, O₂
viridissima, O₂, 6
vulgaris, O₂

VERMES

Platyhelminthes *Flatworms*

Dendrocoelum lacteum, βM_2 , 16
Planaria alpina, O₂
gonocephala, O₂, 3, 15

Polycelis cornuta, O₂
nigra, βM_2
Vorticaria pictus, βM_2 ; O₂

Nemathelminthes *Roundworms*

Ancylostoma duodenale, αM_2
Anguillula species, M₁₂
vulgaris, αM_{15} ; βM_{15}
Diplogaster rivalis, αM_2
Gordius aquaticus, O₂
Monhystera species, M₁₂
macrura, αM_2 ; βM_2

Plectus species, M₁₂
tenuis, αM_2
Rhabditis species, M₁₂
Trilobus gracilis, αM_2
Tripyla setifera, αM_2 , 9

Trochelminthes *Trocal Worms*

For Rotifera see p. 550.

Gastrotricha

Chaetonotus species, M₁₂
maximus, βM_2 ; O₂
larus, βM_2 ; O₂
Dasydytes longisetosum, βM_2

Dasydytes saltitans, βM_2
zelinkai, βM_2
Ichthyidium podura, O₂
Lepidoderma rhomboides, βM_2

Coelhelminthes *Annelida or Segmented Worms*

Aelosoma quaternium, αM_2
Chaetogaster diaphanus, βM_2 ; O₂
Dero limosa, αM_2
Enchytraeus humiculator, αM_2
Hæmopsis sanguisuga, βM_2
Haplotaxis gordioides, O₂, 3
Helobdella nepheloides, P₁₈
stagnalis, P₁₈
Helodrilus tetraedrurus, βM_2
Herpobdella atomaria, βM_{15}
punctata, P₁₈
Limnodrilus hoffmeisteri, P₁₈; αM_2
udekemianus, αM_2

Lumbricillus lineatus, αM_2 , 15
Lumbriculus variegatus, αM_2 ; βM_2
Lumbricus rubellus, αM_2
Nais species, αM_2
elinguis, αM_2 ; βM_2 , 15
Nephelis vulgaris, αM_2 , 3; βM_2 , 3, 9; O₂
Phreorytes menkeanus, O₂, 3
Stylaria lacustris, βM_2 , 3, 15
Tubifex species, P₁₅; αM_{15} ; βM_{15}
tubifex, P₂, 3, 6, 10; αM_6 , 9, 12; M₂, 3

ARTHROPODA

Arachnida Water Spiders, Water Mites and Water Bears

Arrhenurus bicuspidator, βM_2
Atax crassipes, O_2
Curvipes nodatus, O_2
 rufus, O_2
Hydrachna species, M_{17} ; O_2
 globosa, βM_2

Hygrobatas nigro-maculatus, O_2
Limnesia maculata, βM_2
Limnochares holosericeus, O_2
Macrobiotus caconyx, βM_2
Neumania spinipes, O_2
Tardigrada group, M_{12} , 16

Insecta

Acilius sulcatus, O_2 , 3
 * *Aeschna grandis*, O_2
Agabus bipustulatus, O_2
 * *Agrion puella*, O_2
 * *Anabolia laevis*, βM_2
 * *Anax junius*, O_{10}
 * *Baetis* species, O_2
 * *Brachycentrus subnubilus*, βM_2 ; O_2
 * *Cænis fumosa*, αM_2
 * *Calopteryx virgo*, βM_2 ; O_2
 * *Ceratopogon* species, βM_2
 * *Chironomus* (yellow species), βM_2 , 6
 (red species), P_2
 species, P_{15} ; αM_{15}
 crassicauda, M_3
 decorus, βM_{10} ; M_3 , 13
 digitatus, O_3
 dux, O_3
 ferrugineovitlatus, O_3
 frequens, M_3
 lobiferus, M_3
 maturus, M_3
 modestus, O_3
 motitator, αM_2 ; βM_2
 nigricans, O_3
 plumosus, P_3 , 3; αM_2 , 6, 3; βM_2 ; M_3 , 3
 tentans, O_3
 viridicollis, M_3
 * *Cloeon dipterum*, O_2
Colymbetus fuscus, O_2
 * *Corethra plumiconis*, O_3 , 3
Corixa species, O_{15}
 striata, O_2
 * *Corydalis cornuta*, O_{10}
 * *Cricotopus* species, O_3
 trifaciatu, O_3

* *Culex* species, βM_2
 annulatus, βM_2
Dytiscus marginalis, M_2 ; O_2 , 6
 * *Ephemera* species, O_{15}
 vulgata, O_2
 * *Erioptera* species, P_{10}
 * *Eristalis tenax*, P_3 , 3, 6, 10, 16; M_2
Gyrinus natator, O_2
 * *Heptagenia* species, O_{10}
 fluminum, O_2
Hydrometra lacustris, O_2 (Indifferent)
 rufoscutellata, O_2 (Indifferent)
Hydrophilus piceus, O_2
 * *Hydropsyche* species, βM_2 ; O_{10}
 angustipennis, βM_2
 * *Hydroptila sparsa*, O_2
 * *Leptocercus annulicornis*, O_2
 * *Libellula* species, O_{15}
 depressa, O_2
Limnobates stagnorum, O_2 (Indifferent)
 * *Molanna angustata*, βM_2
 * *Nemura variegata*, O_2
Nepa cinerea, O_3 , 15
Notonecta glauca, O_3 , 15
 * *Orthocladius* species, O_3
 * *Oxyethira costalis*, βM_2
 * *Palpomyia* species, O_3
 longipennis, O_3
 * *Perla* species, O_9 , 10
 bicaudata, O_2
 nubecula, O_2
 * *Phryganea grandis*, βM_2 ; O_3 , 9
 striata, O_2 , 3
 * *Polymitarcys virgo*, O_2
 * *Procladius concinnus*, O_3
 culiciformis, O_3

* *Present only in developmental stages as larvæ or pupæ, not present as imago.*

- Prosopistoma foliaceum*, O₂
Psephenus leonti, O₁₀
 * *Psychoda* species, αM_3 ; M₁₂
 alternata, P₁₀
 phalanoides, αM_2 , 6
 sexpunctata, αM_2 , 6
 * *Ptychoptera contaminata*, αM_2
Ranatra linearis, βM_2 ; O₂
 * *Rhyacophila vulgaris*, O₂
 * *Sericostoma* species, βM_2 ; O₂
 * *Sialis lutaria*, αM_2
 * *Simulium ornatum*, αM_2 ; βM_2
 reptans, αM_2 ; βM_2
 * *Stratiomys* species, αM_3
 chamæleon, αM_2
Tæniopteryx trifasciata, O₂
 * *Tanypus dyari*, M₃, 18
 illinoensis, O₈
 monilis, αM_2 ; βM_2 ; O₈
Velia currens, αM_2 (Indifferent)

Mollusca

- Amphipeplea glutinosa*, O₂
Ancylus lacustris, βM_2 ; O₂
 fluviatilis, βM_2 ; O₂, 16
Anodonta mutabilis, O₂ (Indifferent)
Aplexa hypnorum, βM_2 ; O₂
Calyculina lacustris, βM_2
Campeloma subdolum, αM_3 ; βM_{18}
Dreissensia polymorpha, O₂, 3, 9, 16
Limnæa auricularia, βM_2 , 3
 ovata, βM_2 , 15
 palustris, βM_2 ; O₂
 peregra, βM_2 ; O₂, 6
 stagnalis, O₂, 3, 6, 9
Lithoglyphus naticoides, βM_2
Margaritana margaritifera, O₂
Musculium transversum, M₃, 18
 truncatum, M₁₈
Neritina fluviatilis, βM_2
Physa acuta, βM_2 ; O₂
 fontinalis, βM_2 , 15; O₂, 15
Pisidium amnicum, O₂
 compressum, M₁₈
 fossarinum, O₂
 pauperculum, M₁₈
Planorbis carinatus, O₂
 corneus, βM_2 ; O₂
 marginatus, O₂
Sphærium corneum, αM_2 , 3, 15; βM_2 , 15
 monanum, βM_2 ; O₂
 rivicolum, βM_2 , 3; O₂, 3
 stamineum, M₁₈
 striatinum, M₁₈
Unio batavus, O₂
 pictorum, O₂ (Indifferent)
 tumidus, βM_2
Valvata piscinalis, βM_2 , 16
 tricarinata, O₁₈
Viviparus contectus, αM_2 ; βM_2 , 9
 fasciatus, βM_2 , 15

VERTEBRATA

Pisces Fish

- Abramis brama*, O₂
Acerina cernua, O₂
Alburnus lucidus, βM_2 ; M₃; O₃
Anguilla vulgaris, βM_2
Blicca björkna, O₂
Carassius carassius, βM_2 (Indifferent)
 vulgaris, βM_2
Catostomus commersonii, βM_{10}
Cobitis fossilis, βM_2
Cyprinus carpio, βM_2
Esox lucius, O₂
Gasterosteus aculeatus, βM_2 , 3, 6
 pungitius, O₂
Gobio fluviatilis, O₂, 6
Idus melanotus, O₂
Leucaspis delineatus, βM_2
Leuciscus rutilus, O₂
Lota vulgaris, O₂
Lucioperca sandra, O₂
Micropterus dolomieu, O₁₀
 salmoides, O₁₀
Perca fluviatilis, O₁

* *Present only in developmental stages as larvæ or pupæ; not present as imago.*

Rhodeus amarus, βM_2
Salmo sala-sebago, O_{10}
Salvelinus fontinalis, O_{10}

Scardinius erythrophthalmus, O_2
Tinca vulgaris, $\beta M_2, 3$
Trutta fario, $O_2, 3$

Amphibia

Rana fusca, βM_2 (Eggs and tadpoles in-
different)
esculenta, βM_2 (Eggs and tadpoles in-
different)

Triton cristatus, O_2
tæniatus, O_2

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GLOSSARY OF SCIENTIFIC TERMS

acerate, needle-shaped.
acicular, slender or needle-shaped.
adoral, pertaining to the mouth.
æruiginous, blue-green in color.
agglutinate, glued together.
Akinetes, single cells whose walls thicken and separate from the thallus.
ala (æ), a wing-like organ or process.
Algicide, a substance destructive to algae.
amoeboid, like an amoeba.
amorphous, formless.
amylaceous, starchy.
Anabolism, constructive metabolism.
anal, relating to the anus.
analytic, resolving into constituent parts.
androgynous, having the characters of both male and female.
Annulus (i), a ring.
Antenna (æ), a slender process suggesting the "feelers" of insects.
Antennule, a small antenna.
Antheridium (ia), an organ in which male reproductive cells are produced.
Antherozoid, male motile ciliated cells produced in the antheridium.
antibiotic, antipathetic, antagonistic.
apical, at the apex or tip.
Aplanogametes, non-ciliated gametes.
Aplanospore, a non-motile resting spore for tiding over unfavorable conditions.
oppose, placed opposite or before.
arborescent, tree-like.
Archegone, an organ in which female reproductive cells are produced.
arcuate, bent like a bow, curved.
articulate, jointed.
Ascus (i), a spore sac of certain fungi in which spores are formed.
attenuate, narrowed, tapered.
Auricle, a small lobe or ear.
auto-, a prefix meaning *self* or *independent*.

Auxospore, a diatom that has been reduced to a minimum size by successive reproduction, that escapes from its valve by bi-partition, that unites with a similar cell and that thus gives rise to a new plant.
axillary, pertaining to the angle formed between the axis and any process which arises from it.
Azygospore, a parthogenetic spore.
bacillar, **bacilliform**, rod-like.
Basidium (ia), a spore cell of certain fungi which forms spores by abstriction.
basipetal, towards the base.
Benthos, an assemblage of bottom organisms.
bi-, a prefix meaning *twice*.
bifid, two-cleft.
binary, consisting of two members.
biramus, two-branched.
Blepharoplast, a granule giving rise to the flagellum.
Branchia (iæ), gills.
branchiform, like gills.
buccal, pertaining to the mouth.
campanulate, bell-shaped.
Canaliculus (i), a small canal.
capitate, enlarged at the tip.
Capsule, a sac-like membrane.
Carapace, a hard shell.
cardioid, heart-shaped.
carinate, keel-shaped.
carpogenic, fruit-producing.
Carpospore, a spore formed at the end of filaments.
caudal, pertaining to the tail.
Cellulin, modification of cellulose, a substance composing the cell wall.
centric, in the middle.
cervical, pertaining to the neck.
Chitin, a horny substance.

- Chlamydospore**, a spore with thick membrane.
- Chlorophyll**, the green coloring matter of plants.
- Chloroplast**, a chlorophyll-carrying body.
- Chromatophore**, a collective term for the various color-carrying bodies in cells.
- Cilium** (ia), a vibratile hair-like process.
- Cingulum** (a), a girdle.
- circinate**, spirally curled or coiled.
- cirrose**, bearing or resembling a tendril.
- Cirrus** (i), a tendril.
- clathrate**, perforated or latticed like a window.
- clavate**, club-shaped.
- Cloaca** (æ), the common chamber into which the intestinal and genital canals open.
- Cœnobium** (ia), a colony of independent organisms in a common investment.
- cœnocyctic**, of the nature of an aggregation of protoplasmic units enclosed in a common wall.
- Cœcœcium** (ia), a common ground work of a colony.
- concatenate**, linked as in a chain.
- confervoid**, composed of threads.
- Conidium** (ia), a fungal spore asexually produced.
- Conjugation**, union or fusion of two gametes to form a zygote.
- contractile**, capable of contracting.
- convolute**, rolled together.
- cordate**, heart-shaped.
- Corm**, a bulb-like stem.
- Corona** (æ), the ciliated disk of certain animals.
- Coronula** (æ), a small corona.
- Cortex**, an enveloping coat or layer.
- corticate**, pertaining to the cortex.
- Costa**, a rib.
- costate**, ribbed.
- crenulated**, provided with small teeth or notches.
- cruciate**, cross-shaped.
- Cuirass**, plates or scales resembling a piece of armor.
- cuneate**, wedge-shaped.
- Cuticle**, the outermost covering.
- cuticular**, pertaining to the cuticle.
- Cyclosis**, circulation of protoplasm within the cell.
- cymbiform**, boat-shaped.
- cymose**, pertaining to or bearing a cell cluster of centrifugal or determinate type.
- Cytoplasm**, the general protoplasm of the cell, excluding the nucleus.
- denticulate**, minutely toothed.
- dermal**, pertaining to the outer covering.
- Diathermancy**, permeation by radiant heat.
- dichotomous**, forked.
- diclinous**, unisexual.
- diffuent**, dissolving.
- diffuse**, spreading.
- dicœcious**, having the sexes separate.
- disciform**, discoid, disk-shaped.
- distal**, standing far apart.
- dorsal**, pertaining to the back.
- Ecology**, the mutual relation between organisms and their environment.
- ecto-**, a prefix meaning *outside* or *outer*.
- emarginate**, notched, usually at the tip.
- encuirassed**, provided with a cuirass.
- encysted**, enclosed in a bag, coat, or capsule.
- endo-**, a prefix meaning *inside* or *inner*.
- Endosome**, a body in the nucleus, a nucleolus.
- entire**, with even margin.
- epi-**, a prefix meaning *above*, *over* or *upon*.
- Epilimnion**, the upper or circulating water strata in a lake.
- eu-**, a prefix meaning *true* or *exclusively*.
- euglenoid**, like *Euglena*.
- evanescent**, vanishing.
- evert**, to turn inside out.
- extensile**, extensible.
- Fascicle**, a tuft, cluster, or bundle.
- fasciculate**, drawn into a fascicle.
- Fetch**, the clear sweep of the wind.
- Fibrillum** (a), a small fiber.
- filiform**, thread-like.

Flagellum, a lash-like process.

Flame cells, terminal cells appearing like a flame.

foliaceous, leaf-like.

foraminal, pitted or marked with little holes.

-form, a suffix meaning *shaped like*.

Fossa (æ), a ditch or trench-like depression.

Frond, a leafy branch.

funicular, consisting of a small cord or band.

furcate, forked.

fusiform, spindle-shaped.

Gamete, a unisexual cell producing another individual after conjugation with another cell to form a zygote.

Ganglion (ia), a mass of nervous matter.

geminate, in pairs.

Gemma (æ), a bud.

Gemmation, bud formation.

Gemmule, one of the internal buds of the porifera.

geniculate, bent like a knee joint.

Germination, budding, beginning to grow.

globose, nearly spherical.

Gonidium (ia), a propagative cell, asexually produced.

granulose, composed of grains.

helio-, a combining form from the Greek word for *the sun*.

heliophilous, sun- or light-loving (photophilous).

heliotactic, moving towards the sun or light (phototactic).

Heliotaxis, motion towards the sun or light (phototaxis).

holo-, a combining form from the Greek word for *whole*.

holophytic, wholly plant-like in nutrition.

holozoic, wholly animal in nutrition.

hydro-, a combining form from the Greek word for *water*.

hyper-, a prefix signifying *over* or *above*.

hypo-, a prefix signifying *under* or *beneath*.

Hypolimnion, the stagnant zone of water in a lake.

illoricate, without a lorica.

incised, with deeply notched margin.

indurated, hardened.

Integument, a covering.

intercalary, between others, interspersed.

involute, rolled inwards.

iso-, a prefix indicating equality.

isogamous, having like gametes.

Isotherm, a line of equal temperature.

Katabolism, destructive metabolism.

limnetic, pertaining to the open water of lakes.

Limnology, the study of lakes.

littoral, pertaining to the shore.

lophophore, an organ bearing tentacles.

lorica, a protective external case.

macro-, a combining form signifying *large*.

Mandible, an anterior pair of mouth organs.

masticatory, adapted for chewing.

Mastax, the gizzard of rotifers.

Matrix, the lifeless tissue between cells.

Maxilla, the jaw.

Membranell, a membrane.

membranous, like a fine membrane.

Mesentery, a peritoneal fold.

meso-, a combining form denoting *in the middle* or *intermediate*.

Mesolimnion, the thermocline or layer of water between the epi- and hypolimnion.

mesosaprobic, intermediately saprobic.

meta-, a prefix meaning *between* or *with*.

metabiotic, symbiotic, with one organism preparing the way for the other.

Metabolism, the process of assimilating and excreting food substances.

metameric, segmented.

Metamorphosis, change of form.

Metazoön (oa), a multicellular animal.

micro-, a combining form signifying *small*.

Microgonidium, a small gonidium.

moniliform, like a necklace or string of beads.

mono-, a prefix signifying *one* or *alone*.

monœcious, with male and female organs in one individual.

-morphic, a combining form meaning *shaped like*.

mucilaginous, like mucilage.

Mucus, a viscid or slimy fluid.

mucronate, ending in a sharp point.

Mycelium, a network of filamentous cells in fungi.

Nannoplankton, dwarf plankton.

Nares (posterior), openings of the olfactory organ into the pharynx or throat.

Nauplius, the earliest larval stage of the entomostraca.

naviculoid, boat-shaped.

Nekton, an assemblage of free-swimming organisms.

nuclease, provided with a nucleus.

Nucleolus (i), a small mass inside the nucleus.

Nucleus (ei), a complex spheroidal mass in the cell.

ob-, a prefix meaning *oppositely* or *inversely*.

ochraceous, sheath-like.

Œsophagus, alimentary canal between pharynx and stomach.

-oid, a suffix meaning *like*.

oligosaprobic, slightly saprobic.

Oöcyte, a stage of the supposedly female protozoan conjugant before it prepares for fertilization.

Oögonium, the mother egg cell.

Oösphere, a female gamete.

Oöspore, the fertilized egg cell.

oral, pertaining to the mouth.

orbicular, circular in outline.

Ovarium (ia), an ovary.

ovate, egg-shaped.

Pabulum, nutriment or food.

Palmella, a zoögeal stage.

Papilla (æ), a nipple-like projection.

Paramylum, a mucilaginous substance probably akin to starch.

Parasite, an organism subsisting on another.

parenchymatous, spongy, porous.

parietal, belonging to the wall.

parthogenetic, developing without fertilization.

pectinate, comb-like.

Pedicie, pedicel, a stalk or support.

pelagic, pertaining to the open water.

Pellicle, a delicate superficial membrane.

pellucid, clear, translucent.

penicillate, pencil-shaped.

Periplasm, protoplasm in the oögonium and the antheridium that does not share in conjugation.

Peristome, the region surrounding the mouth.

Pharynx, the gullet.

-philous, a combining form meaning *loving*.

Photo-, a combining form meaning *light*.

Photosynthesis, the formation of complex organic compounds from simple inorganic ones under the stimulus of light.

phreatic, pertaining to the ground.

phylogenetic, pertaining to ancestral history deduced from development.

-phyte, **phyto-**, combining forms signifying *plant*.

-phytic, a combining form signifying *pertaining to plants*.

pinnatifid, feather-shaped.

planktic, pertaining to plankton.

Plankton, an assemblage of drifting organisms.

Plasm, see protoplasm.

-plast, suffix denoting *thing* or *body*.

Plastid, minute granule found in the cell protoplasm.

plicate, folded into plaits.

Podium (ia), a foot or foot-like structure.

poly-, a prefix signifying *many*.

polymorphic, of diverse form.

Polypid, an individual of a colony.

polysaprobic, very saprobic.

Polyzoarium (ia), the skeletal system of a bryozoan colony.

- potamo-**, a combining form signifying *river*.
- prehensile**, adapted to seize.
- Proboscis**, a trunk-like process of the head.
- Proliferation**, proliferous development.
- prostrate**, lying flat, collapsed.
- Proteolysis**, decomposition of proteins.
- Protista**, an assemblage of primitive forms of life.
- proto-**, a combining form signifying *first* or *primitive*.
- Protophytum** (a), the simplest plant form.
- Protoplasm**, living substance.
- Protoplast**, a cell with or without a wall.
- Protozoön** (oa), the simplest animal form.
- pseudo-**, a combining form or prefix meaning *false*.
- Pseudopodium**, a false foot-like support.
- pulvinate**, cushion-like.
- punctate**, dotted.
- Punctum** (a), a dot.
- pyramide**, pyramid-shaped.
- Pyrenoid**, a minute colorless body embedded in the chromatophore.
- pyriform**, pear-shaped.
- quadrate**, squared.
- quaternate**, arranged in fours.
- radiate**, proceeding from a point.
- Ramification**, branching.
- Ramus**, a branching structure.
- Raphé**, a seam-like line.
- reniform**, kidney-shaped.
- replicate**, doubled down.
- reticulate**, latticed or netted.
- retractile**, capable of being drawn back.
- rheo-**, a combining form signifying *stream*.
- rhizoid**, root-like.
- Rostrum**, a beak-like process.
- Rotule**, a radially-directed bar.
- rudimentary**, imperfectly developed or in an early stage of development.
- saccate**, pouched.
- saprobic**, growing in water containing decomposing organic matter.
- Saprophyte**, an organism (plant) living on dead and decaying matter.
- Sarcode**, body protoplasm of protozoa.
- scalariform**, marked like a ladder.
- scrobiculate**, marked with minute depressions, pitted.
- Seiche**, a synchronous rising and falling of the water in lakes.
- Septum** (a), a partition.
- seriate**, arranged in series or rows.
- serrated**, notched like a saw.
- sessile**, sitting or attached.
- Seta** (æ), a bristle-like structure.
- setiferous**, bristle-bearing.
- setiform**, like a bristle.
- sigmoid**, S-shaped.
- sinuate**, with a wavy margin.
- Sperm**, **Spermatium** (a), a male reproductive cell.
- spicate**, spiked.
- Spicule**, a small needle-shaped body.
- spinous**, having spines.
- spinulose**, having minute spines.
- Sporangiospore**, a sporangium spore.
- Sporangium** (a), a capsule or sac in which spores are produced.
- Spore**, a cell which is freed and can develop into a new individual.
- Sporocarp**, the covering or capsule enclosing a spore.
- Sporulation**, spore formation.
- Statoblast**, a bud or winter egg of the bryozoa and porifera.
- Stauros**, the central nodule of the diatom cell, or a transverse base without markings.
- stellate**, star-shaped.
- Stigma** (ata), a colored spot.
- Stipe**, a support or stem.
- Stipule**, a stem.
- Stria** (æ), a furrow.
- Stylet**, a small pointed bristle-like appendage.
- sub-**, a prefix signifying *under* or *below*.
- suctorial**, adapted for sucking.
- suture**, a line or seam of junction, a union.
- Swarm spore**, a motile protoplasmic body, or a zoöspore.
- symbiotic**, living together with benefit to one or both.

synthetic, combining elements to make a compound.

tabular, flattened.

tactile, sensitive to touch.

taxonomic, classifying.

Tegument, a cover or covering.

Tentacle, a slender process sensitive to touch.

tesselated, checkered.

Test, the shell or hardened outer covering of crustacea.

tetra-, a combining form signifying *four*.

Tetrad, a body of four cells.

Thallus (1), a vegetative body without differentiation into stem and leaf.

Thermocline, a region of sharp change in temperature forming a dividing layer between the circulating and the stagnant water.

Thorax, the body region below or behind the head.

torulose, having small swellings.

Trichite, **Trichocyst**, a fine rod- or needle-like protrusible structure found in certain protozoa.

Trichogyne, an elongated hair-like receptive cell found in certain algæ.

Trichome, a hair-like process, the thread or filament of certain algæ.

Trochus, the inner, anterior, coarser ciliary zone of a rotifer disk.

tuberculate, having small swellings or humps.

tubiculous, having a tube.

turbinate, shaped like a top.

undulate, wavy.

uni-, a prefix signifying *one* or *once*.

utriculate, inflated, bladder-like.

Vacuole, a cavity in the cell plasm.

vegetative, growing.

Velum, a veil-like structure.

ventral, pertaining to the belly.

verrucose, warty.

verticillate, whorled.

vesiculiform, bladder-like or vessel-like.

vibratile, capable of motion or vibration.

villous, bearing long weak hairs.

Vitta (æ), a longitudinal rib.

viviparous, germinating while attached to the parent.

Zoary, the whole of a bryozoan colony.

zonate, zoned or marked with rings.

Zoö dendrium, the tree-like branched stalk of certain colonial infusoria.

Zoö gamete, a motile gamete.

Zoö glæa, a viscous jelly-like mass containing living organisms.

Zoö id, a member of a compound animal organism.

-zoön (zoa), a combining form signifying *animal*.

Zoö spore, a motile spore.

Zygospore, a spore resulting from conjugation.

Zygote, see gamete.

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DIATOMACEÆ, BACILLARIEÆ OR DIATOMS

PLATE I

DIATOMACEÆ, BACILLARIEÆ OR DIATOMS

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<i>a</i> , outer, or older valve. <i>b</i> , inner, or younger valve. <i>c</i> , <i>c'</i> , connective bands or girdles. <i>d</i> , central nodule. <i>e</i> , terminal nodules. <i>f</i> , raphé. <i>g</i> , furrows. <i>m</i> , chromatophore plates. <i>n</i> , nucleus. <i>o</i> , oil globules. <i>p</i> , cavities. <i>u</i> , protoplasm.	
<i>D, E, F</i> , sectional views showing multiplication by division. <i>After Deby.</i>	
<i>a</i> , valve. <i>b</i> , girdle. <i>c</i> , protoplasm. <i>d</i> , chromatophore plates. <i>e</i> , central cavities. <i>f</i> , nucleus and nucleolus. <i>g</i> , oil globules.	

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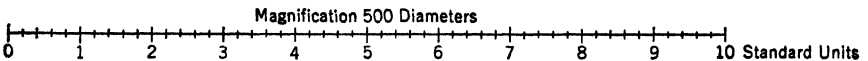


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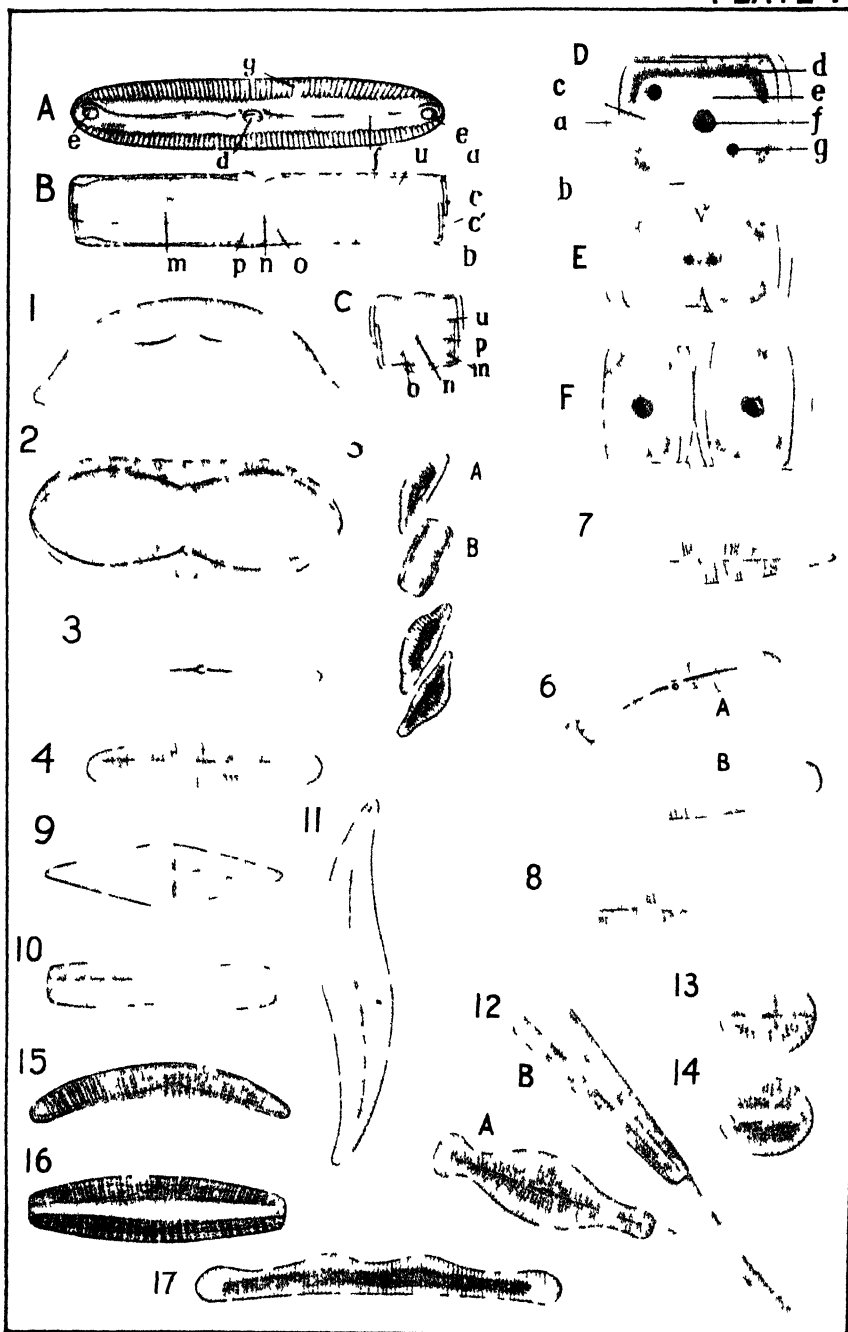


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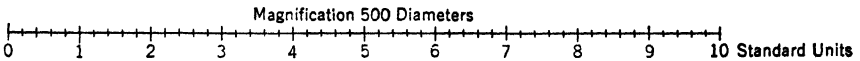
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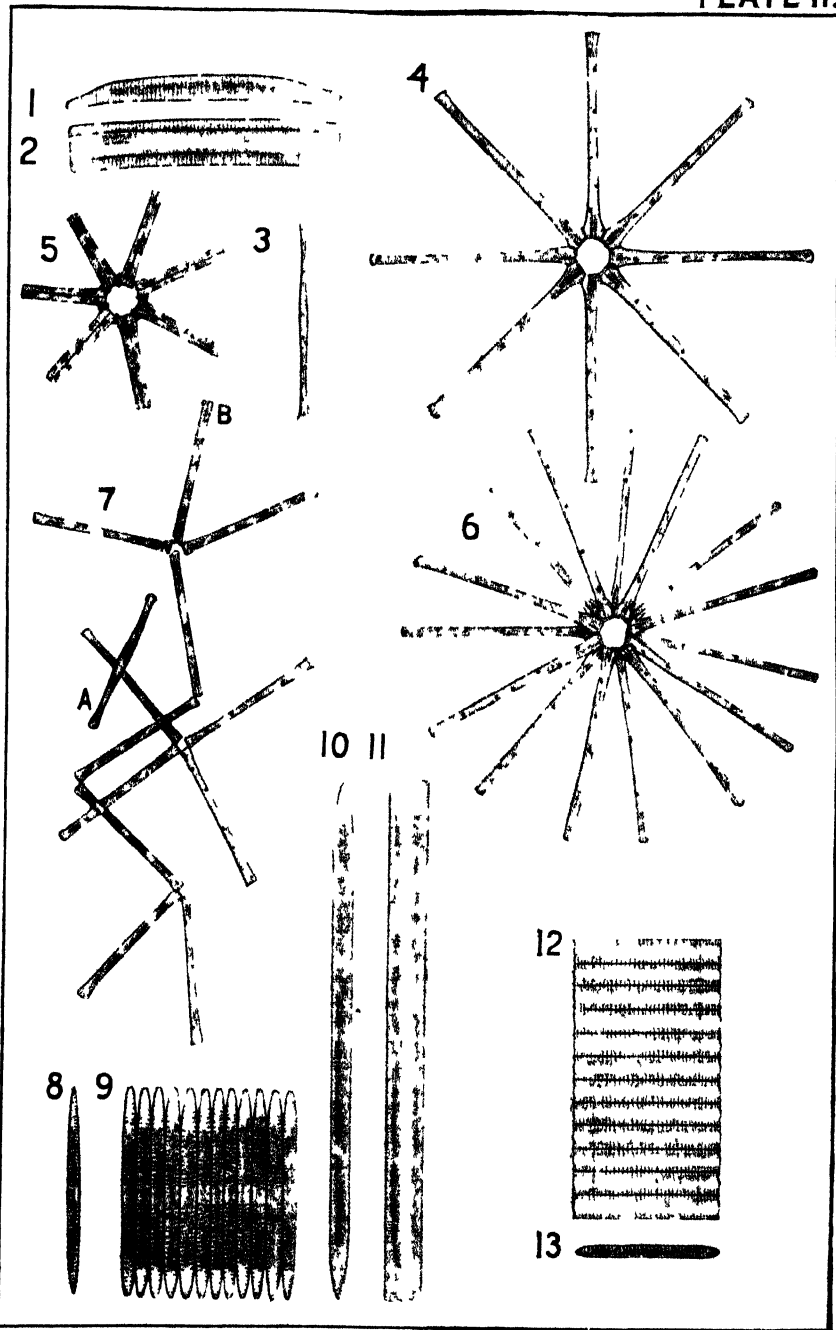


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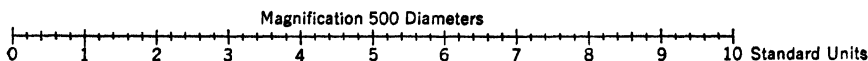
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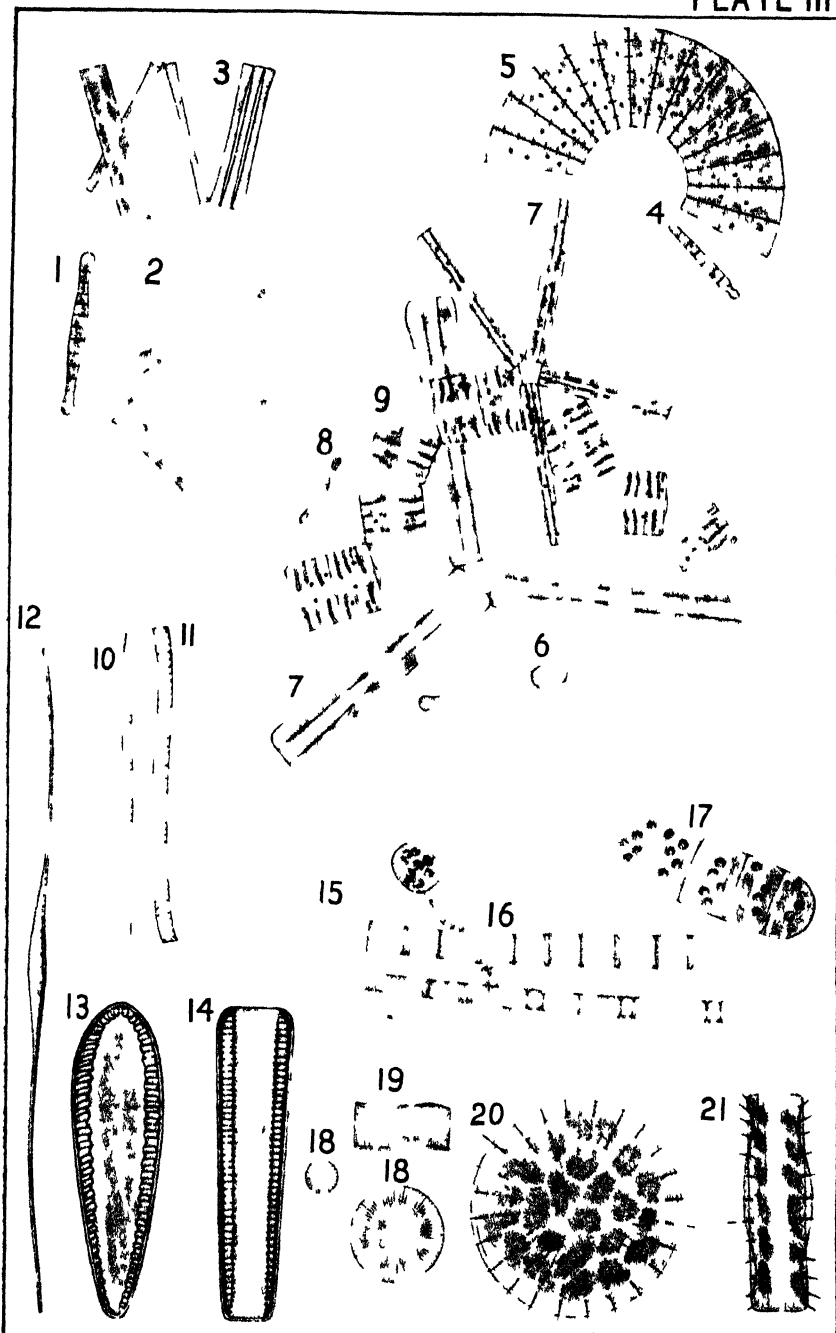


PLATE IV
SCHIZOMYCETES OR FISSION FUNGI
AND
CYANOPHYCEÆ, MYXOPHYCEÆ OR BLUE-GREEN ALGÆ

PLATE IV

SCHIZOMYCETES OR FISSION FUNGI

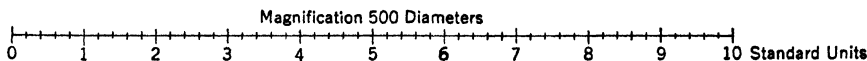
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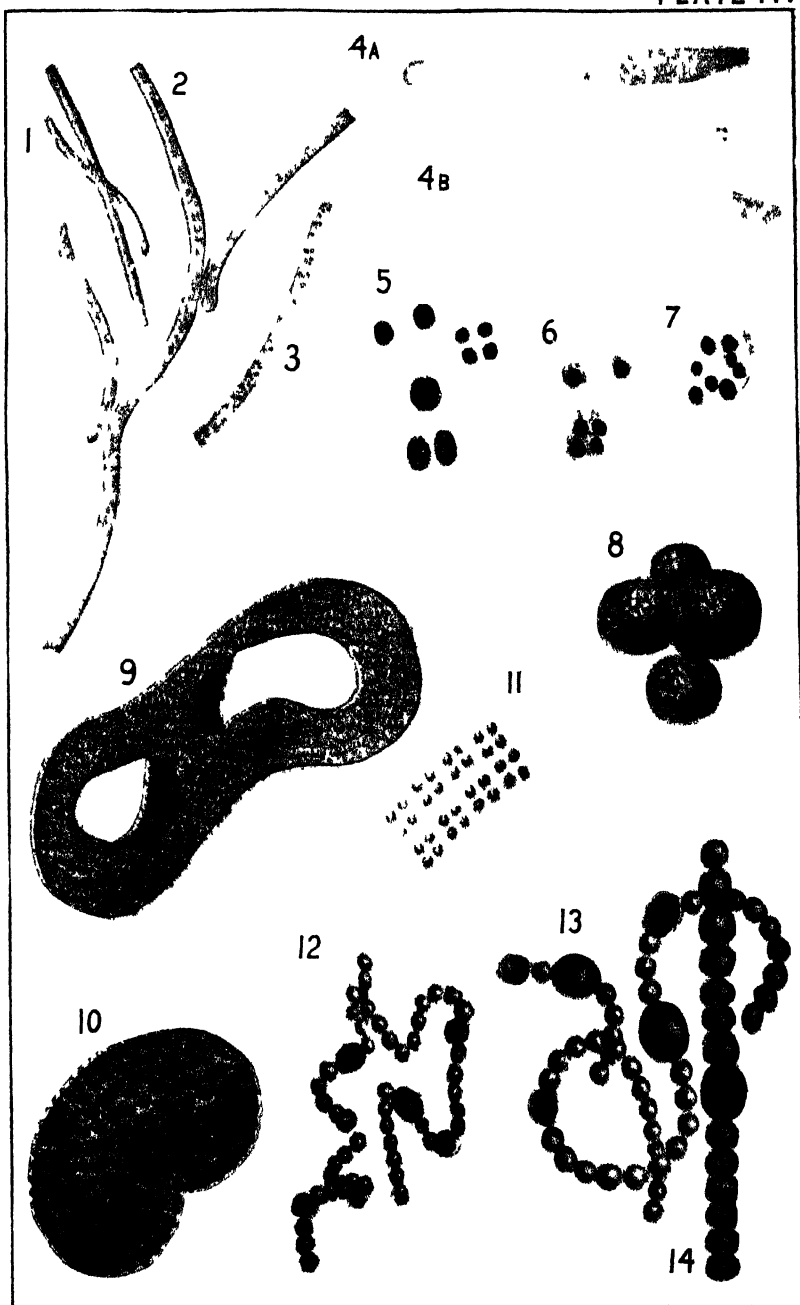


PLATE V

CYANOPHYCEÆ, MYXOPHYCEÆ OR BLUE-GREEN ALGÆ
AND
CHLOROPHYCEÆ OR GREEN ALGÆ

PLATE V

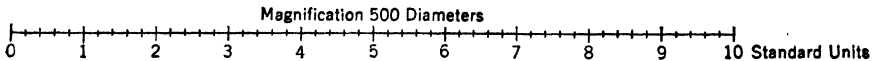
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12. <i>Tetraspora</i>	472

SCALE



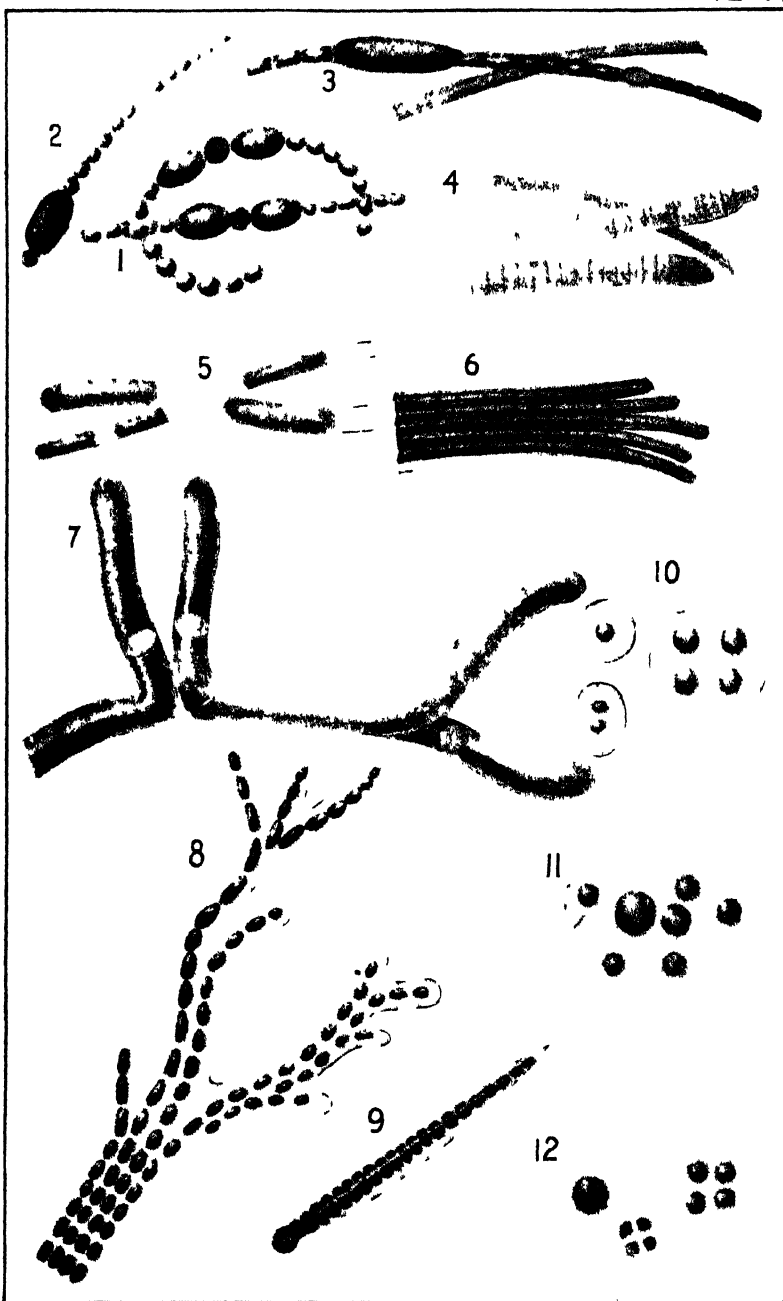


PLATE VI

CHLOROPHYCEÆ OR GREEN ALGÆ

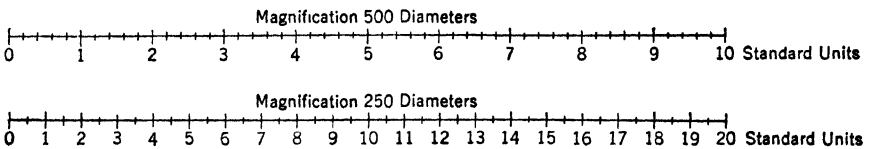
PLATE VI

CHLOROPHYCEÆ OR GREEN ALGÆ

FIGURE	PAGE
¹ 1. Botryococcus. $\times 500$.	483
2. Ankistrodesmus. $\times 500$.	473
3. Dictyosphaerium. $\times 500$.	472
4. Nephrocytium. $\times 500$.	473
5. Dimorphococcus. $\times 500$.	472
6. Protococcus. $\times 500$.	472
7. Tetraëdron. $\times 500$.	473
8. Scenedesmus. $\times 500$.	473
9. Hydrodictyon. $\times 250$.	474
¹ 10. Ophiocytium. $\times 500$.	483
11. Pediatrurn. $\times 500$.	474
12. Sorastrum. $\times 500$.	473

¹Belonging more specifically to the Xanthophyceæ, Heterokontæ or Yellow-Green Algæ.

SCALES



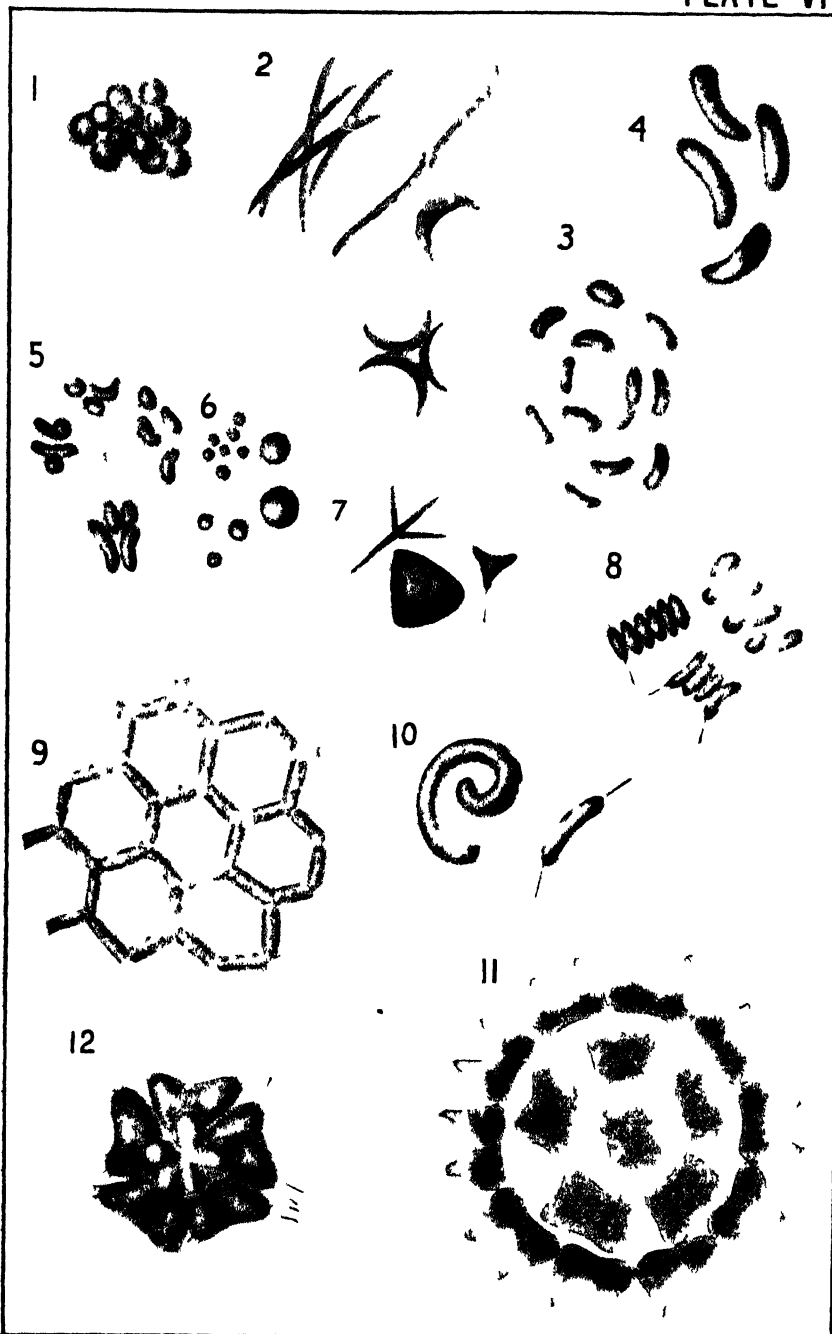


PLATE VII

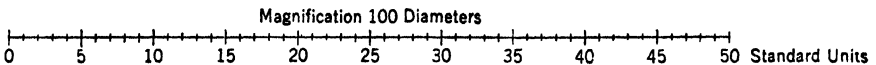
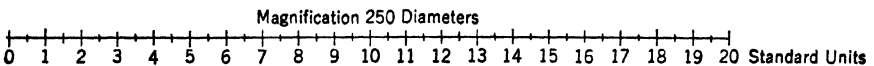
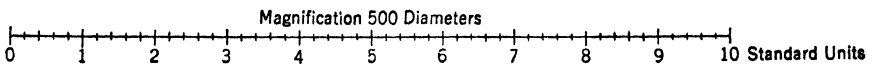
CHLOROPHYCEÆ OR GREEN ALGÆ

PLATE VII

CHLOROPHYCEÆ OR GREEN ALGÆ

FIGURE	PAGE
1. <i>Cœlastrum</i> . $\times 500$	473
2. <i>Crucigenia</i> . $\times 500$	473
3. <i>Volvox</i> . $\times 100$	471
4. <i>Eudorina</i> . $\times 250$	471
5. <i>Pandorina</i> . $\times 250$	470
6. <i>Gonium</i> . <i>a</i> , top view. <i>b</i> , side view. $\times 500$	470
7. <i>Penium</i> . $\times 250$	477
8. <i>Closterium</i> . $\times 250$	477
9. <i>Closterium</i> . $\times 250$	477
10. <i>Closterium</i> . $\times 250$	477
11. <i>Docidium</i> . $\times 250$	477
12. <i>Cosmarium</i> . $\times 250$	477
13. <i>Tetmemorus</i> . $\times 250$	478

SCALES



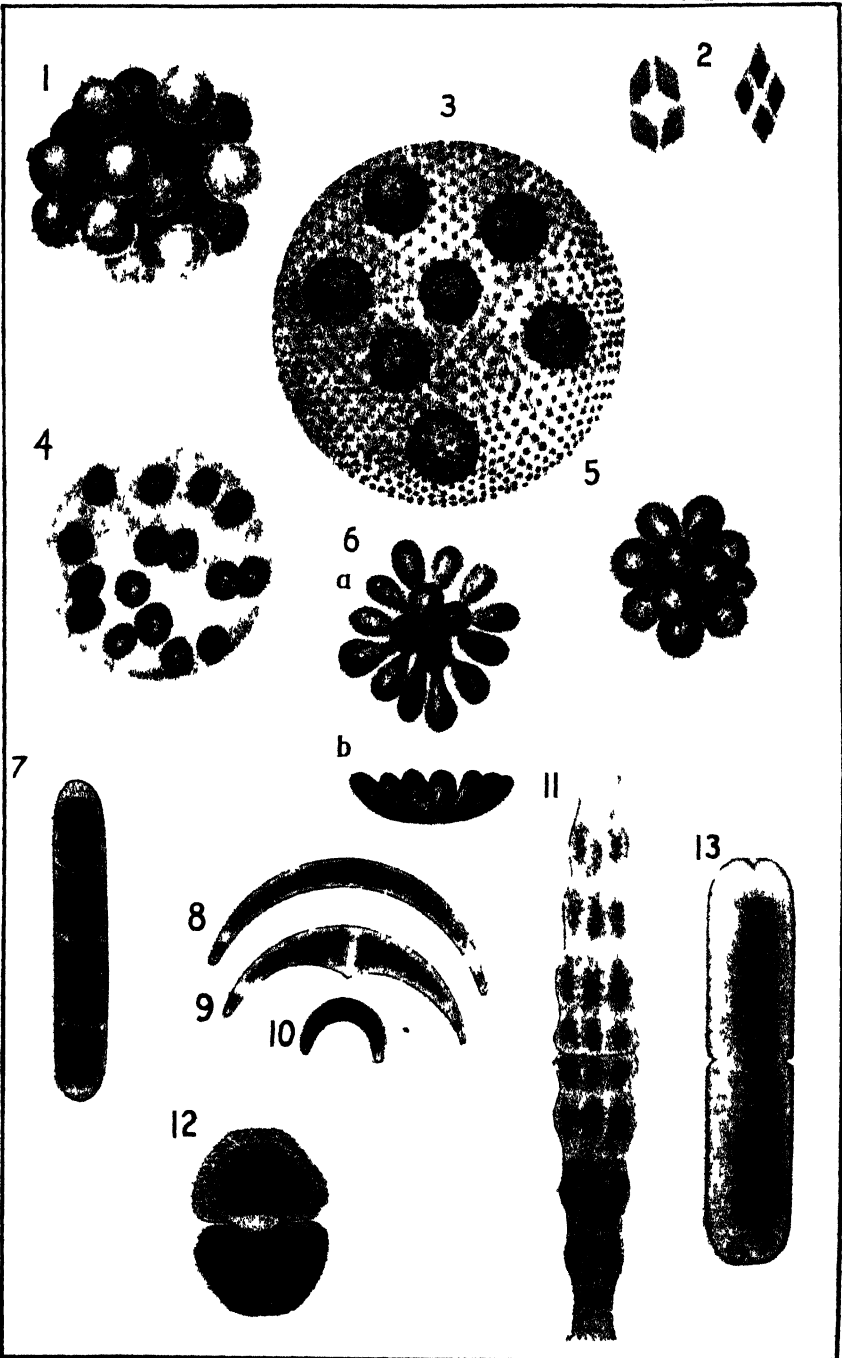


PLATE VIII

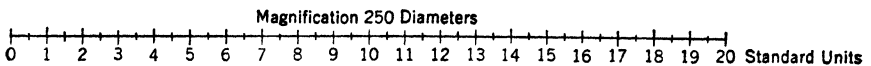
CHLOROPHYCEÆ OR GREEN ALGÆ

PLATE VIII

CHLOROPHYCEÆ OR GREEN ALGÆ

FIGURE	PAGE
1. <i>Xanthidium armatum</i>	478
2. <i>Xanthidium antilopæum</i> . <i>a</i> , front view. <i>b</i> , lateral view. <i>c</i> , end view....	478
3. <i>Arthrodesmus</i> . <i>a</i> , front view. <i>b</i> , end view.....	478
4. <i>Euastrum</i> . <i>a</i> , front view. <i>b</i> , lateral view.....	478
5. <i>Micrasterias</i>	478
6. <i>Staurastrum</i> . <i>a</i> , front view. <i>b</i> , end view.....	478
7. <i>Staurastrum</i> . <i>a</i> , front view. <i>b</i> , end view.....	478
<i>A</i> . <i>Cosmarium</i> , showing division.....	477
<i>B</i> . <i>Cosmarium</i> , showing conjugation.....	477
<i>C, D</i> . <i>Cosmarium</i> , showing formation of zygospore.....	477
<i>E, F</i> . <i>Cosmarium</i> , showing germination of zygospore.....	477

SCALE



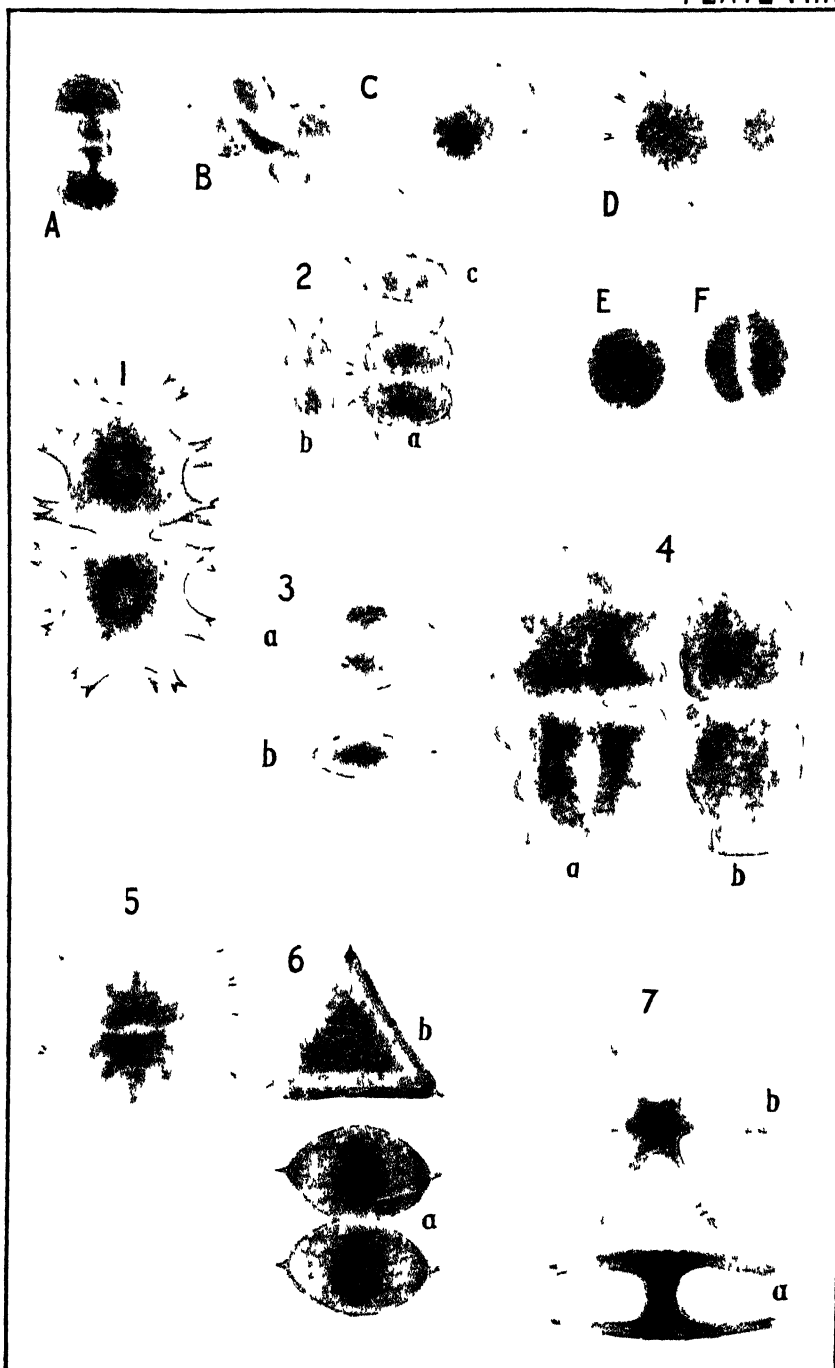


PLATE IX

CHLOROPHYCEÆ OR GREEN ALGÆ

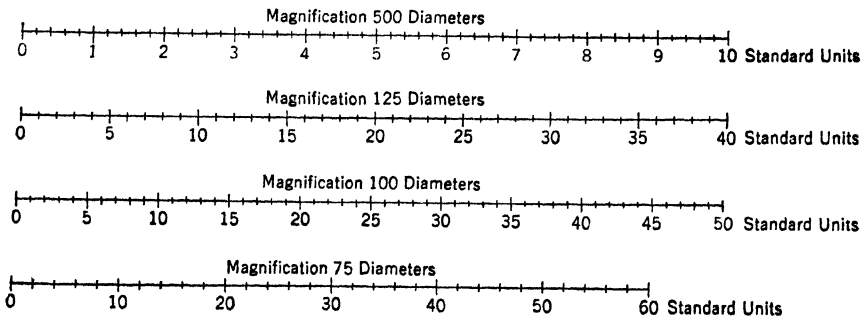
PLATE IX

CHLOROPHYCEÆ OR GREEN ALGÆ

FIGURE	PAGE
1. Hyalotheca. <i>a</i> , filament. <i>b</i> , end view. $\times 500$	478
2. Desmidium. <i>a</i> , filament. <i>b</i> , end view. $\times 500$	478
3. Sphærozozma. <i>a</i> , filament. <i>b</i> , end view. $\times 500$	478
4. Spirogyra. $\times 125$	479
5. Spirogyra, conjugated form, showing spores. $\times 125$	479
6. Zygnema. $\times 125$	479
7. Vaucheria. $\times 100$	475
¹ 8. Tribonema. $\times 125$	483
9. Cladophora. $\times 75$	475
10. Ulothrix. $\times 125$	475

¹Belonging more specifically to the Xanthophyceæ, Heterokontæ or Yellow-Green Algæ.

SCALES



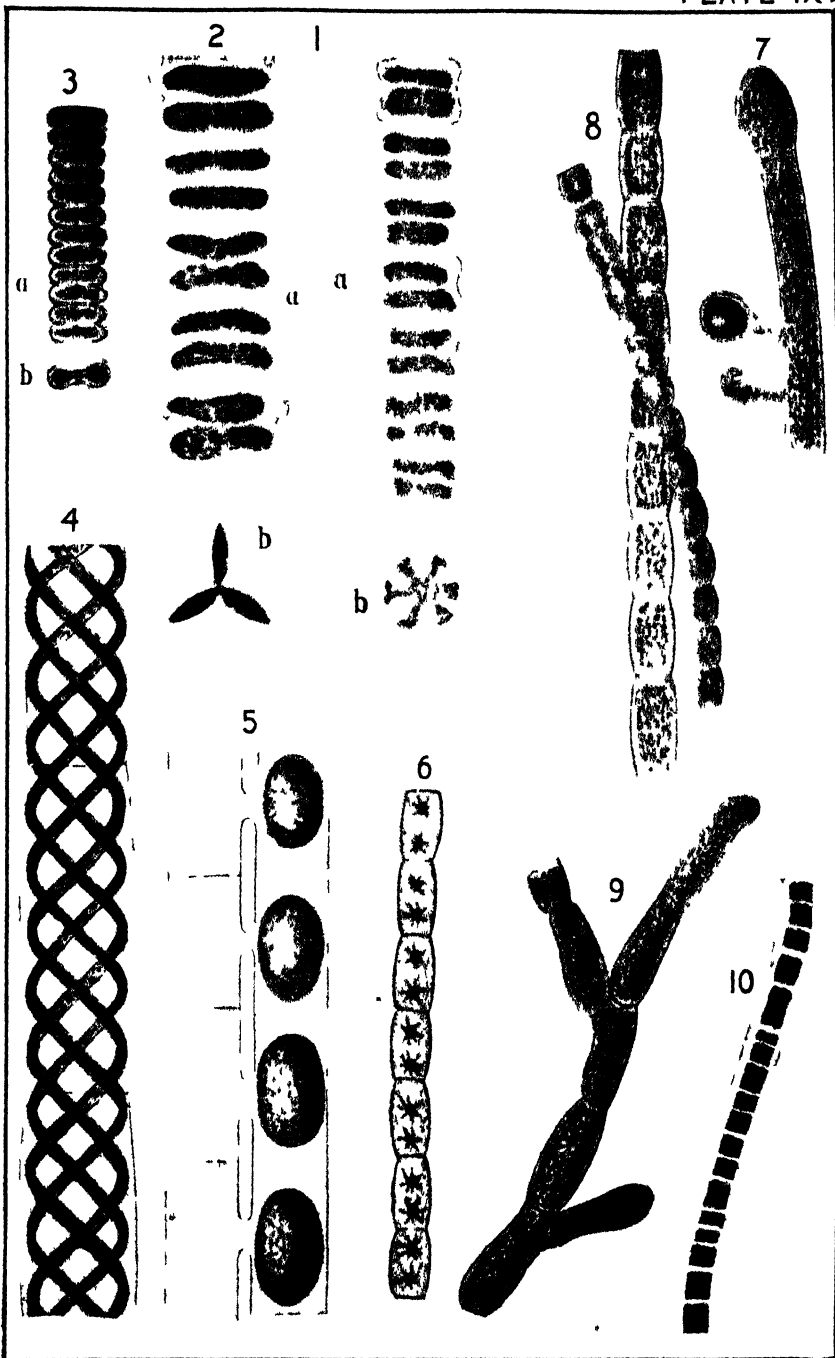


PLATE X

CHLOROPHYCEÆ OR GREEN ALGÆ
AND
MISCELLANEOUS FUNGI

PLATE X

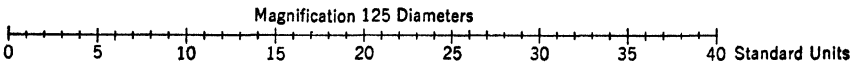
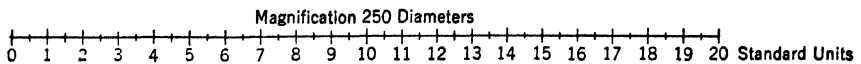
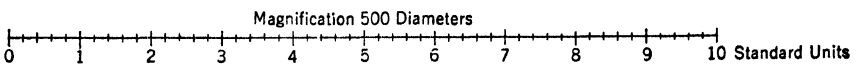
CHLOROPHYCEÆ OR GREEN ALGÆ

FIGURE	PAGE
1. <i>Draparnaldia</i> . $\times 125$	476
2. <i>Stigeoclonium</i> . $\times 125$	476
3. <i>Chætophora</i> . $\times 125$	476

MISCELLANEOUS FUNGI

4. <i>Saccharomyces</i> . $\times 500$. (yeast).....	497
5. Mold hyphæ. $\times 250$	497
6. <i>Penicillium</i> . $\times 250$. (mold).....	497
7. <i>Aspergillus</i> . $\times 250$. (mold).....	497
8. <i>Mucor</i> . $\times 250$. (mold).....	497

SCALES



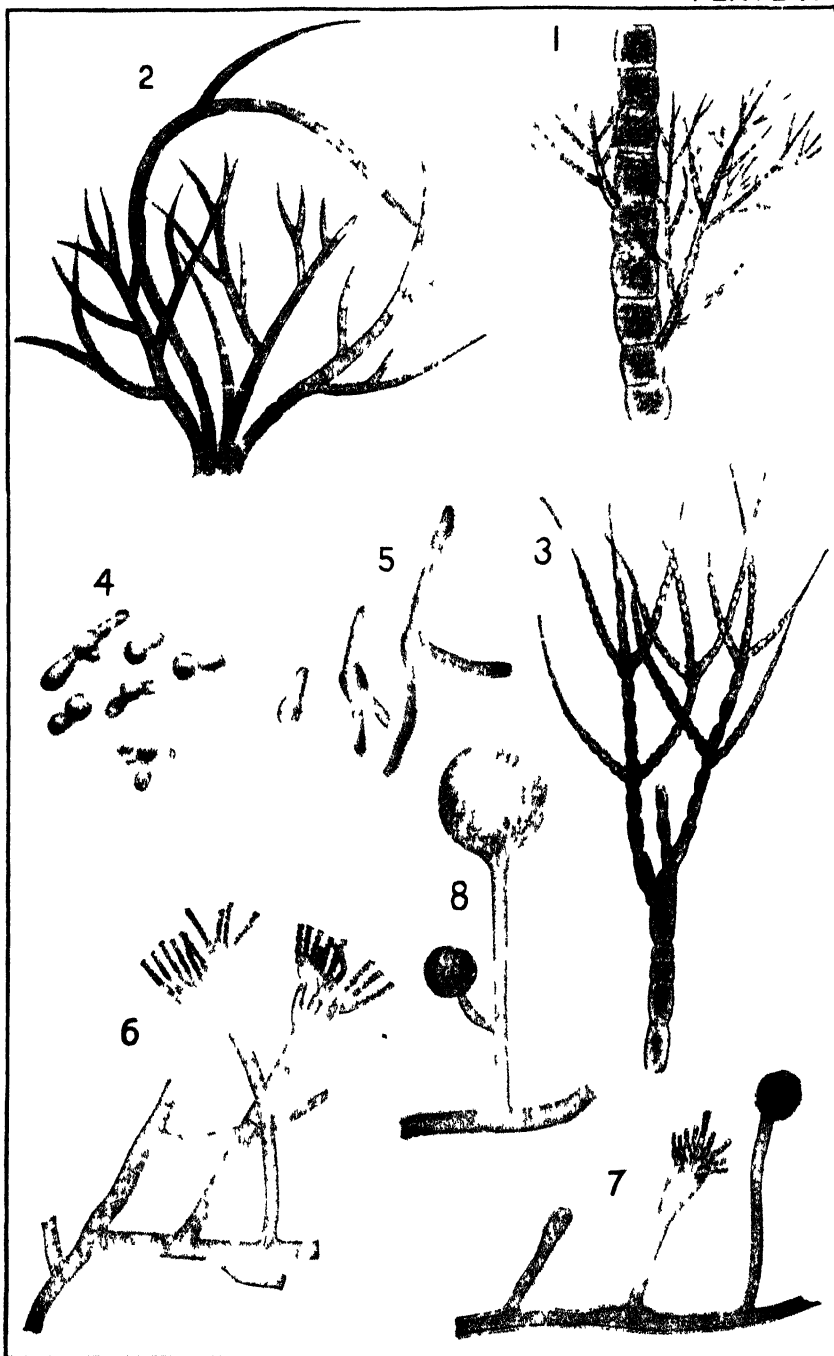


PLATE XI
PHYCOMYCETES
AND
SARCODINA OR AMŒBOID PROTOZOA

PLATE XI

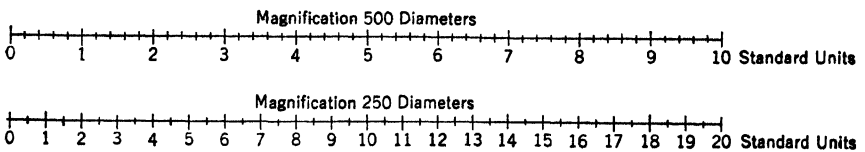
PHYCOMYCETES

FIGURE	PAGE
1. Saprolegnia. $\times 250$	504
2. Achlya. $\times 250$	504
3. Leptomitius (Apodya). $\times 500$	503

SARCODINA OR AMŒBOID PROTOZOA

4. Amœba. $\times 250$	511
5. Arcella, lateral view. $\times 250$	511
6. Arcella, superior view. $\times 250$	511
7. Diffugia. $\times 250$	511
8. Euglypha. $\times 250$	511
9. Trinema. $\times 250$	511
10. Actinophrys. $\times 250$	510

SCALES



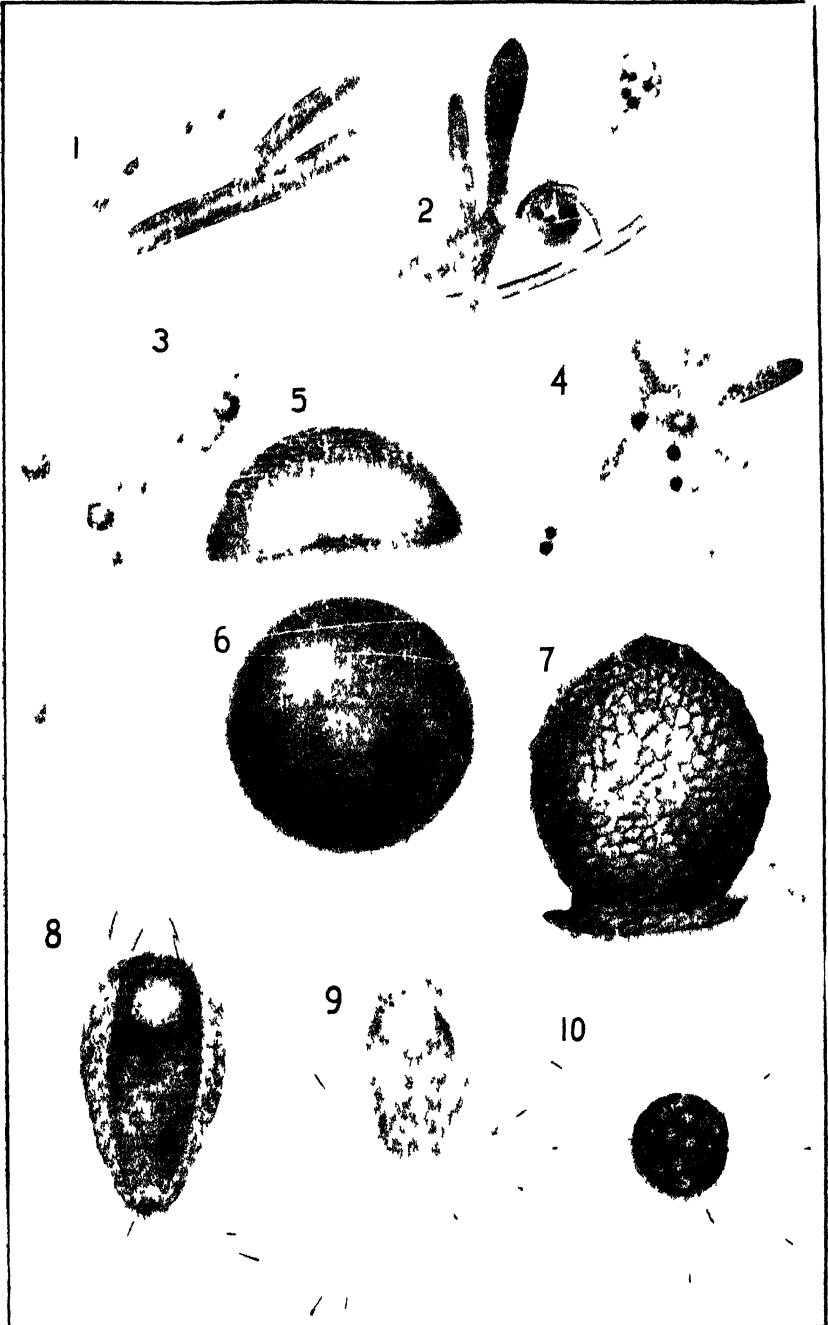


PLATE XII

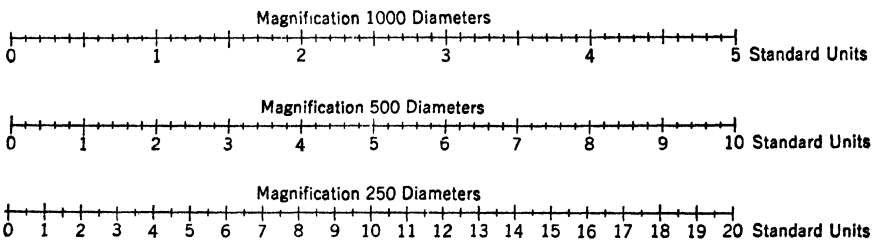
MASTIGOPHORA OR FLAGELLATE PROTOZOA

PLATE XII

MASTIGOPHORA OR FLAGELLATE PROTOZOA

FIGURE	PAGE
1. <i>Cercomonas</i> . $\times 500$	516
2. <i>Monas</i> . $\times 500$	516
3. <i>Anthophysa</i> . $\times 500$	516
4. <i>Vacuolaria</i> . $\times 500$
5. <i>Rhaphidomonas</i> (<i>Gonyostomum</i>). $\times 500$	515
6. <i>Euglena</i> . $\times 500$	515
7. <i>Trachelomonas</i> . $\times 500$	515
8. <i>Phacus</i> . $\times 500$	515
9. <i>Synura</i> . $\times 500$	512
10. <i>Synura</i> . $\times 500$	512
11. <i>Syncritya</i> . $\times 500$	512
12. <i>Uroglenopsis</i> . $\times 250$	513
13. <i>Uroglenopsis</i> , showing division of the monads. $\times 1000$	513

SCALES



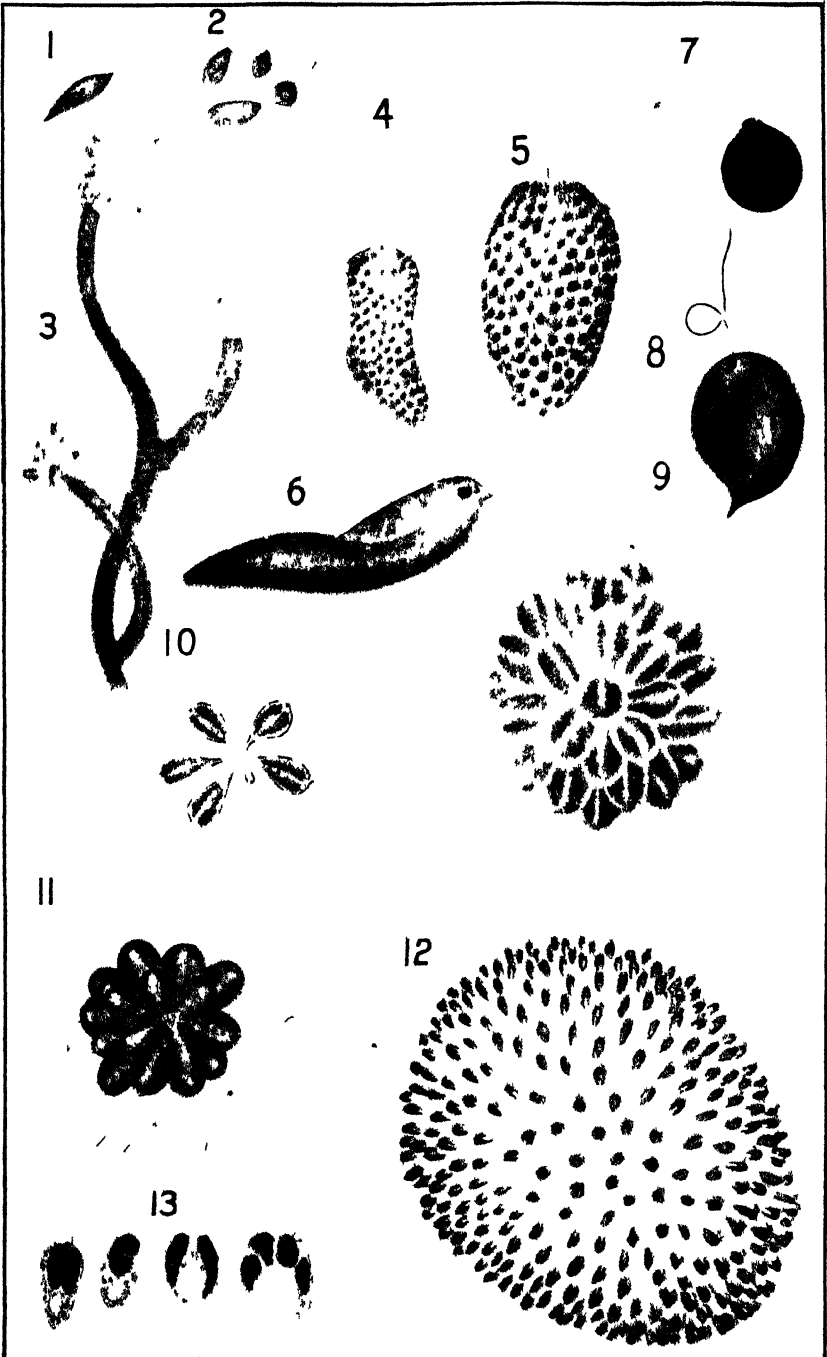


PLATE XIII

**MASTIGOPHORA OR FLAGELLATE PROTOZOA
AND
INFUSORIA OR CILIATE PROTOZOA**

PLATE XIII

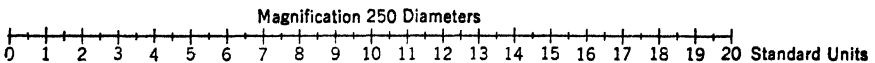
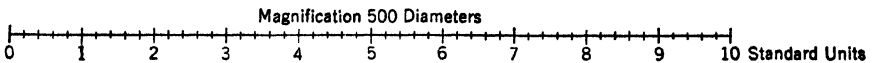
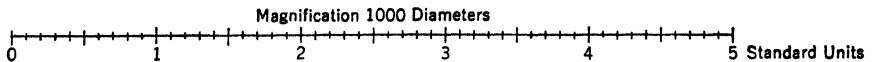
MASTIGOPHORA OR FLAGELLATE PROTOZOA

FIGURE	PAGE
1. Dinobryon. $\times 500$	513
2. Cryptomonas. $\times 500$	513
3. Mallomonas. $\times 500$	512
4. Chlamydomonas. $\times 1000$	514
5. Peridinium. $\times 500$	514
6. Ceratium. $\times 250$	514
7. Glenodinium. $\times 500$	514

INFUSORIA OR CILIATE PROTOZOA

8. Euplotes. $\times 250$	520
9. Halteria. $\times 500$	519
10. Vorticella. $\times 250$	520
11. Epistylis. $\times 250$	520
12. Tintinnus. $\times 250$	519

SCALES



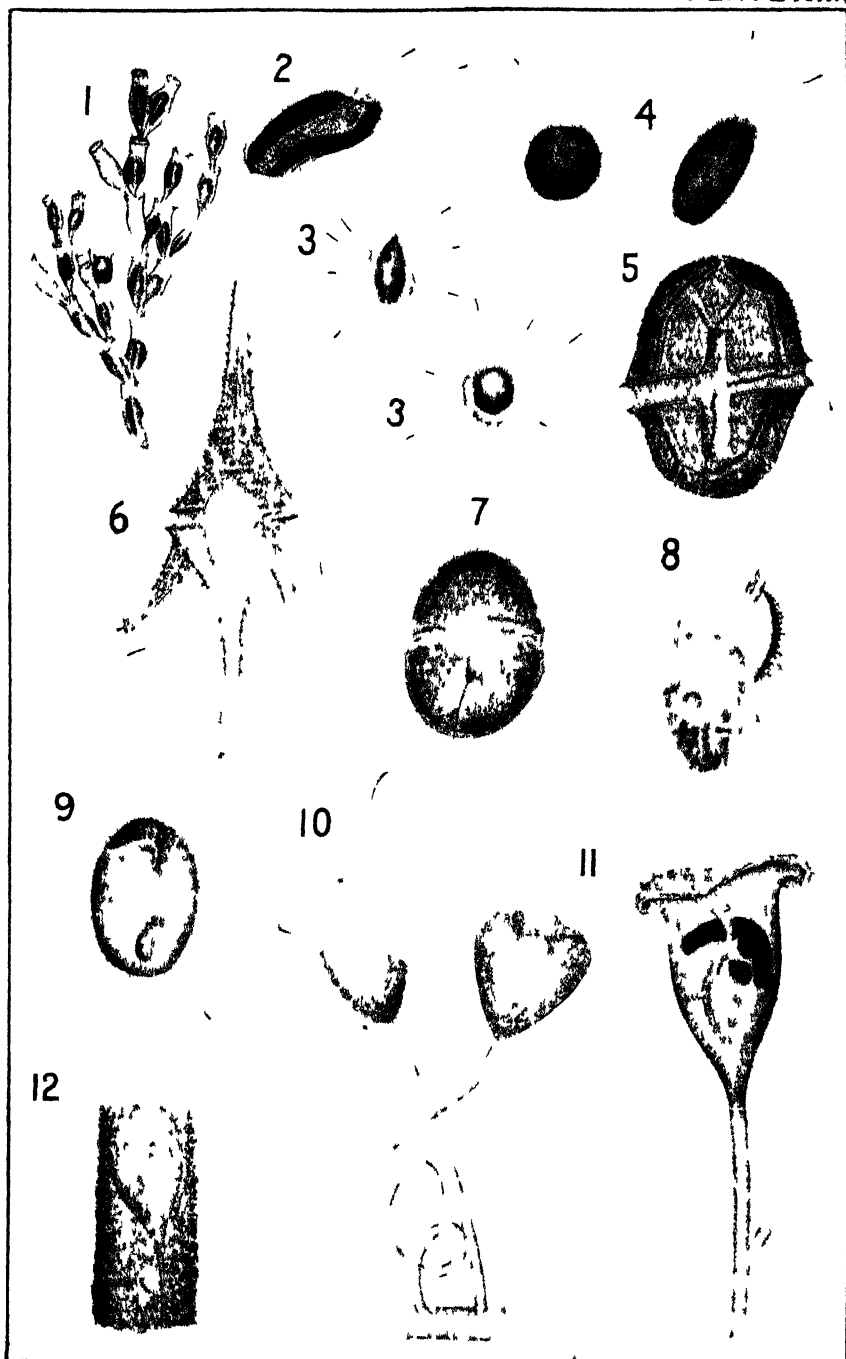


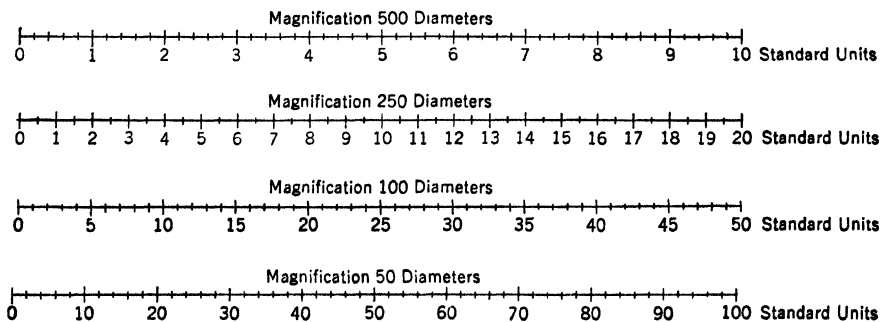
PLATE XIV
INFUSORIA OR CILIATE PROTOZOA

PLATE XIV

INFUSORIA OR CILIATE PROTOZOA

FIGURE	PAGE
1. Codonella. $\times 500$	519
2. Stentor. $\times 50$	518
3. Bursaria. $\times 100$	518
4. Paramecium. $\times 250$	517
5. Nassula. $\times 250$	517
6. Coleps. $\times 500$	517
7. Enchelys. $\times 500$	518
8. Trachelocerca. $\times 500$	518
9. Pleuronema. $\times 500$	518

SCALES



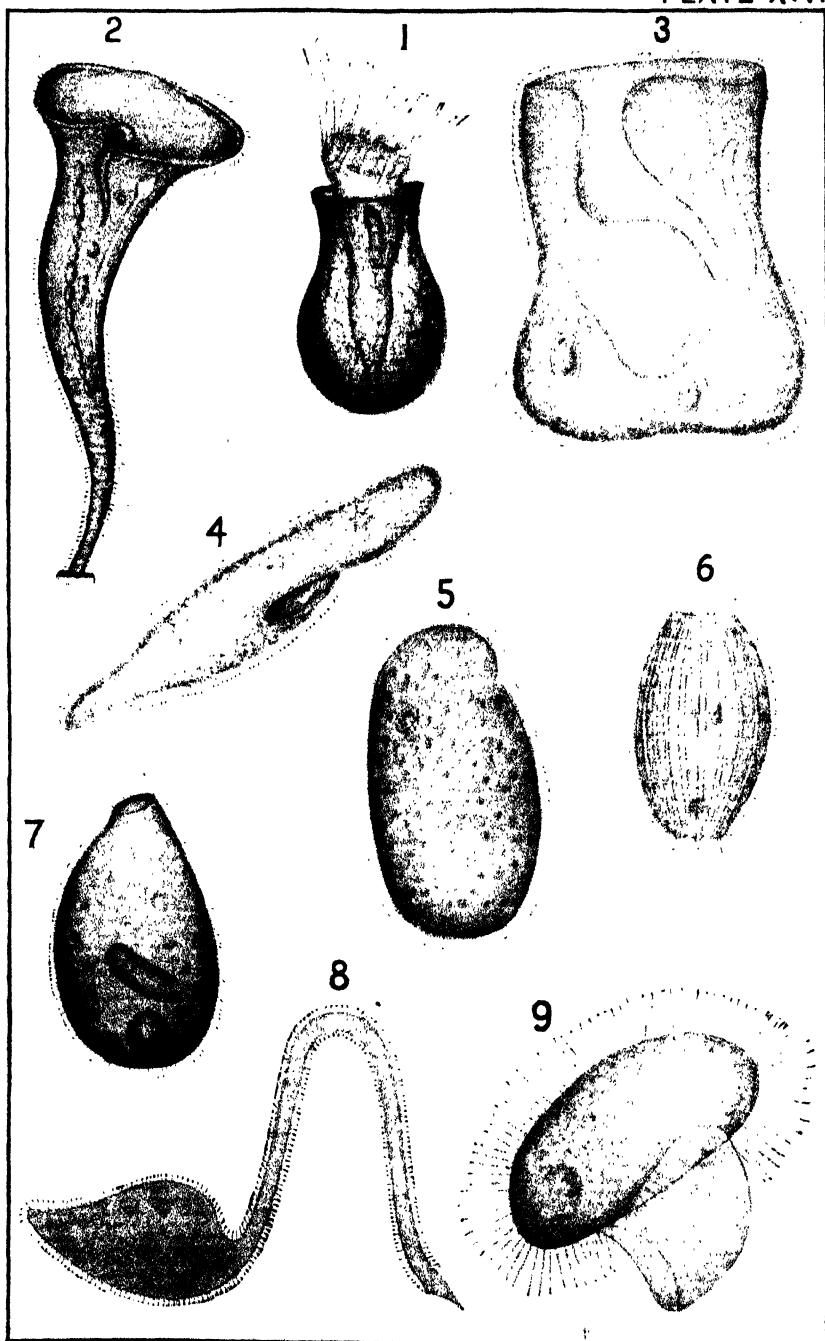


PLATE XV
INFUSORIA OR CILIATE PROTOZOA
AND
ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES

PLATE XV

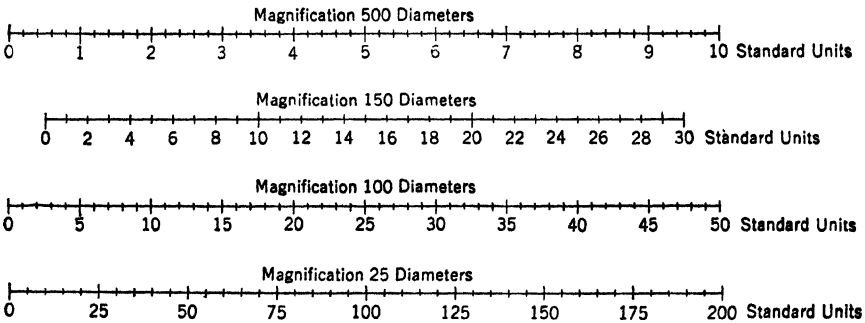
INFUSORIA OR CILIATE PROTOZOA

FIGURE	PAGE
1. Colpidium. $\times 500$	518
2. Acineta. $\times 500$	521

ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES

3. Floscularia. $\times 25$	526
4. Melicerta (Floscularia). $\times 25$	527
5. Conochilus. $\times 100$	527
6. Rotifer. $\times 100$	527
7. Microcodon. $\times 150$
8. Asplanchna. $\times 150$	527

SCALES



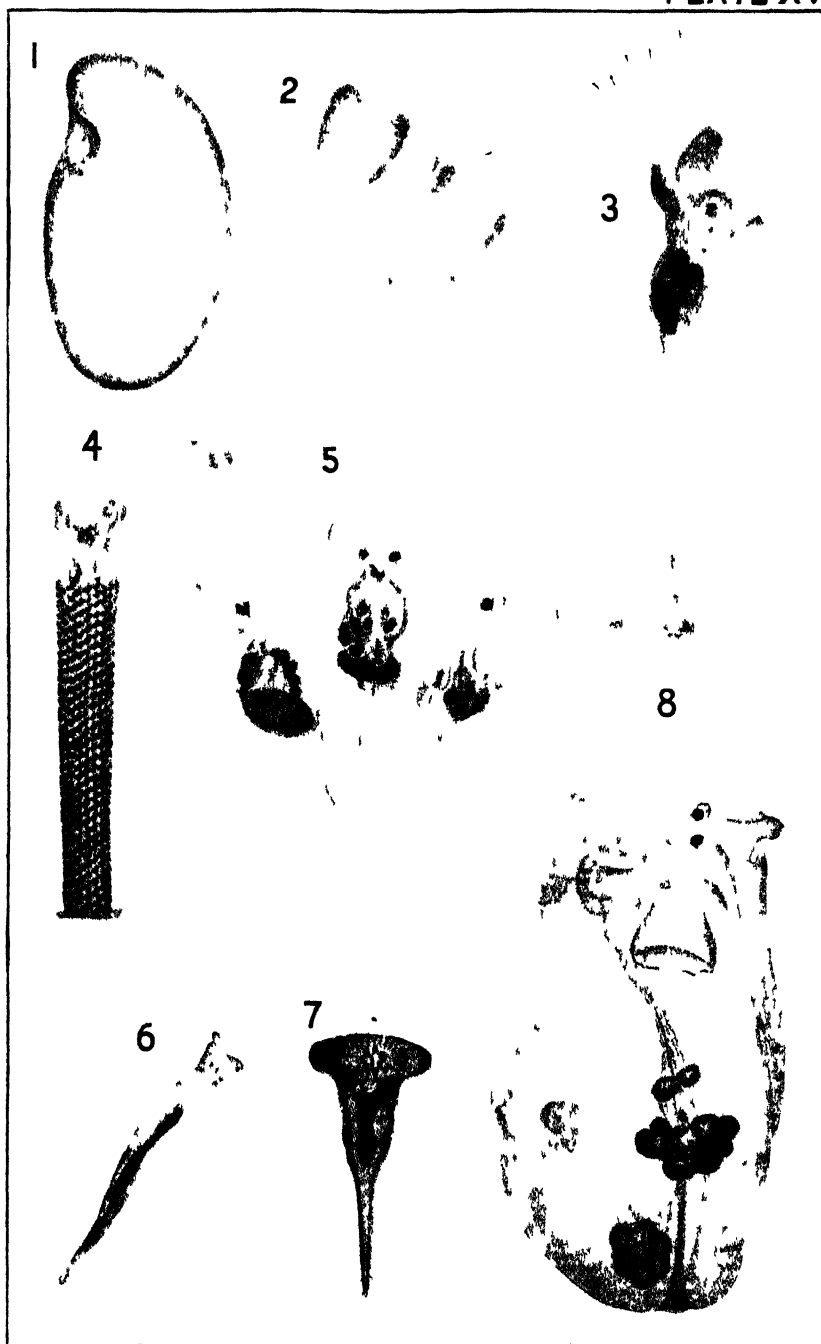


PLATE XVI

ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES

PLATE XVI

ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES

FIGURE	PAGE
1. Synchæta. $\times 100$
2. Polyarthra. $\times 200$	527
3. Triarthra. $\times 150$
4. Diglena (Dicranophorus). $\times 150$	528
5. Tricercera (Mastigocerca, or Rattulus). $\times 150$

TROCHAL DISKS. (*After Bourne*)

A. Microcodon.....	...
B. Stephanoceros.....	...
C. Hypothetical intermediate form.....	...
D. Philodina.....	527
E. Brachionus.....	528

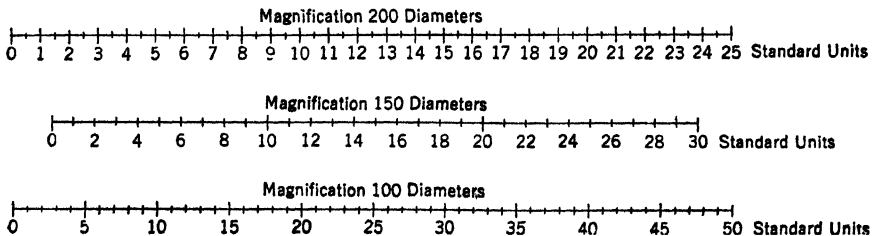
STRUCTURE OF FOOT. (*After Hudson and Gosse*)

F. Floscularia, rhizotic foot.....	526
G. Melicerta (Floscularia), rhizotic foot.....	527
H. Rotifer, bdelloidic foot.....	527
I. Pedalion, scirtopodic foot.....	...

FORMS OF TROPHI. (*After Hudson and Gosse*)

J. malleate; *K.* submalleate; *L.* forcipitate; *M.* incudate; *N.* uncinatæ; *O.* ramate; *P.* malleoramate.

SCALES



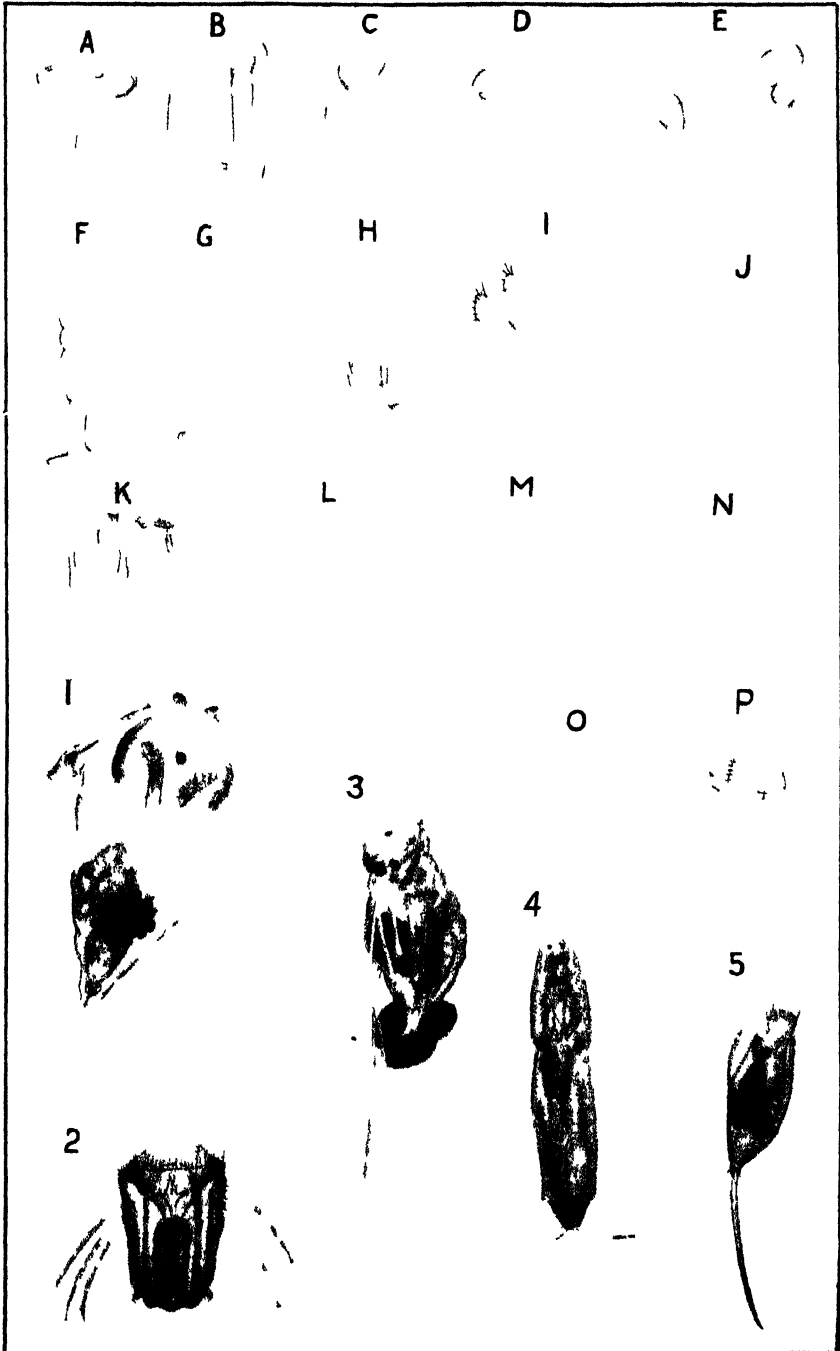


PLATE XVII

**ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES
AND
CRUSTACEA**

PLATE XVII

ROTIFERA, ROTATORIA OR WHEEL ANIMALCULES

FIGURE	PAGE
1. <i>Brachionus</i> . $\times 200$	528
2. <i>Anuræa</i> (<i>Keratella</i>) <i>cochlearis</i> . $\times 150$. <i>A</i> , dorsal view, <i>B</i> , side view.....	528
3. <i>Anuræa aculeata</i> (<i>Keratella quadrata</i>). $\times 150$	528
4. <i>Notholca</i> . $\times 200$

CRUSTACEA

COPEPODA

5. <i>Cyclops</i> . $\times 25$	533
6. <i>Diaptomus</i> . $\times 25$	533
7. <i>Canthocamptus</i> . $\times 25$	533

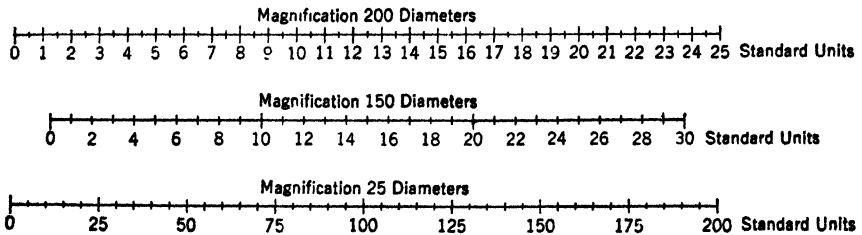
OSTRACODA

8. <i>Cypris</i> . $\times 25$	533
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CLADOCERA OR WATER FLEAS

9. <i>Daphnia</i> . $\times 25$	532
10. <i>Bosmina</i> . $\times 25$	532

SCALES



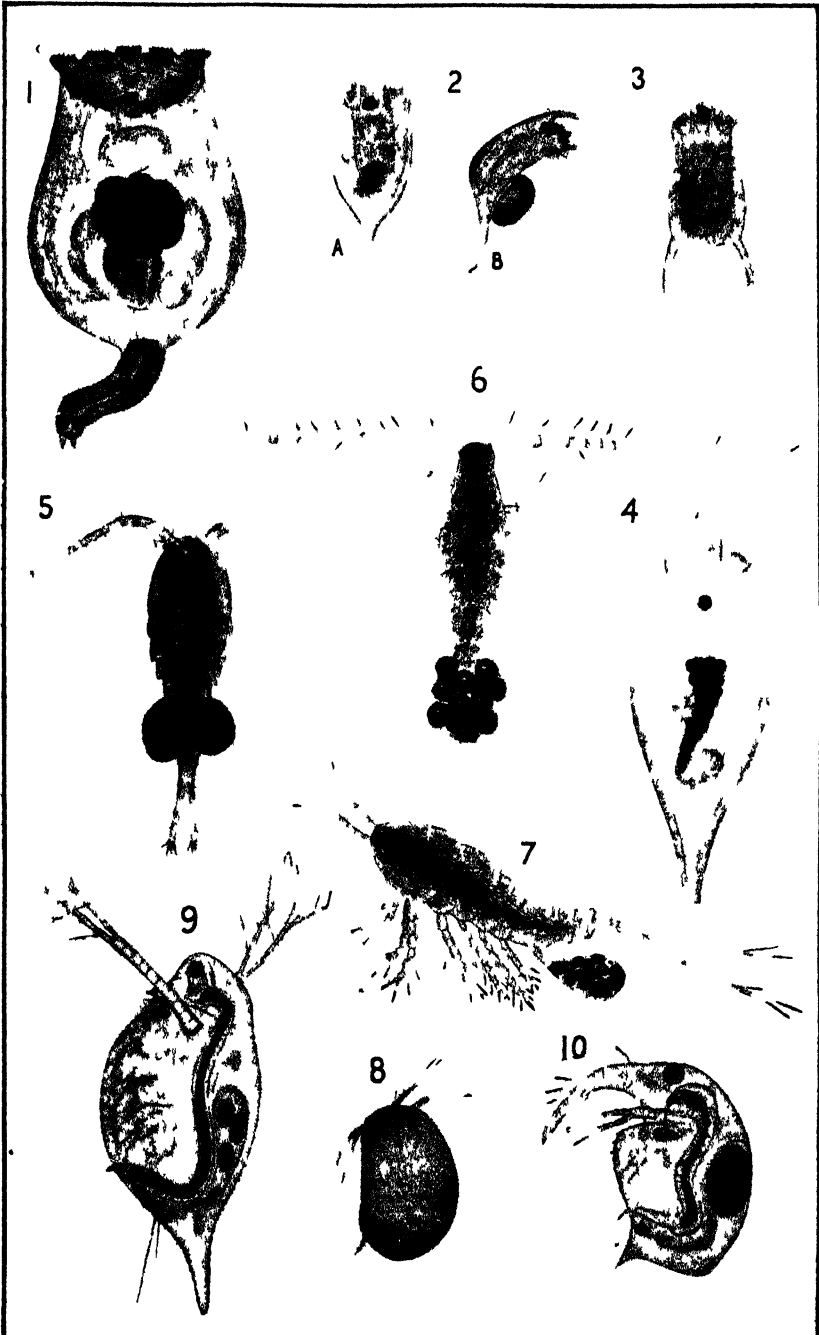


PLATE XVIII

**CRUSTACEA; BRYOZOA OR MOSS ANIMALCULES
AND
PORIFERA OR SPONGES**

PLATE XVIII

CRUSTACEA

CLADOCERA OR WATER FLEAS

FIGURE	PAGE
1. Sida. $\times 25$	532
2. Chydorus. $\times 25$	532

PHYLLOPODA OR FAIRY SHRIMPS

3. Eubranchipus. $\times 2$	531
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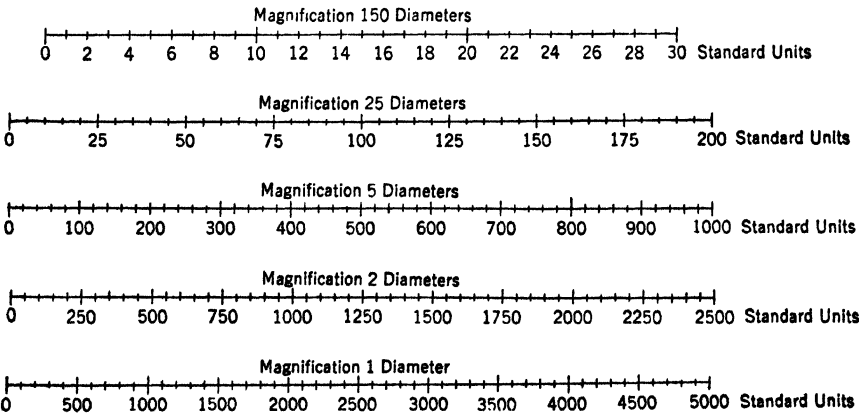
BRYOZOA OR MOSS ANIMALCULES

4. Fredericella. $\times 5$	536
5. Paludicella. $\times 5$	536
6. Plumatella, statoblast. $\times 25$	536
7. Pectinatella, statoblast. $\times 25$	439, 536

PORIFERA OR SPONGES

8. Spongilla. $\times 1$	538
9. Sponge spicules (skeleton spicules). $\times 150$	538

SCALES



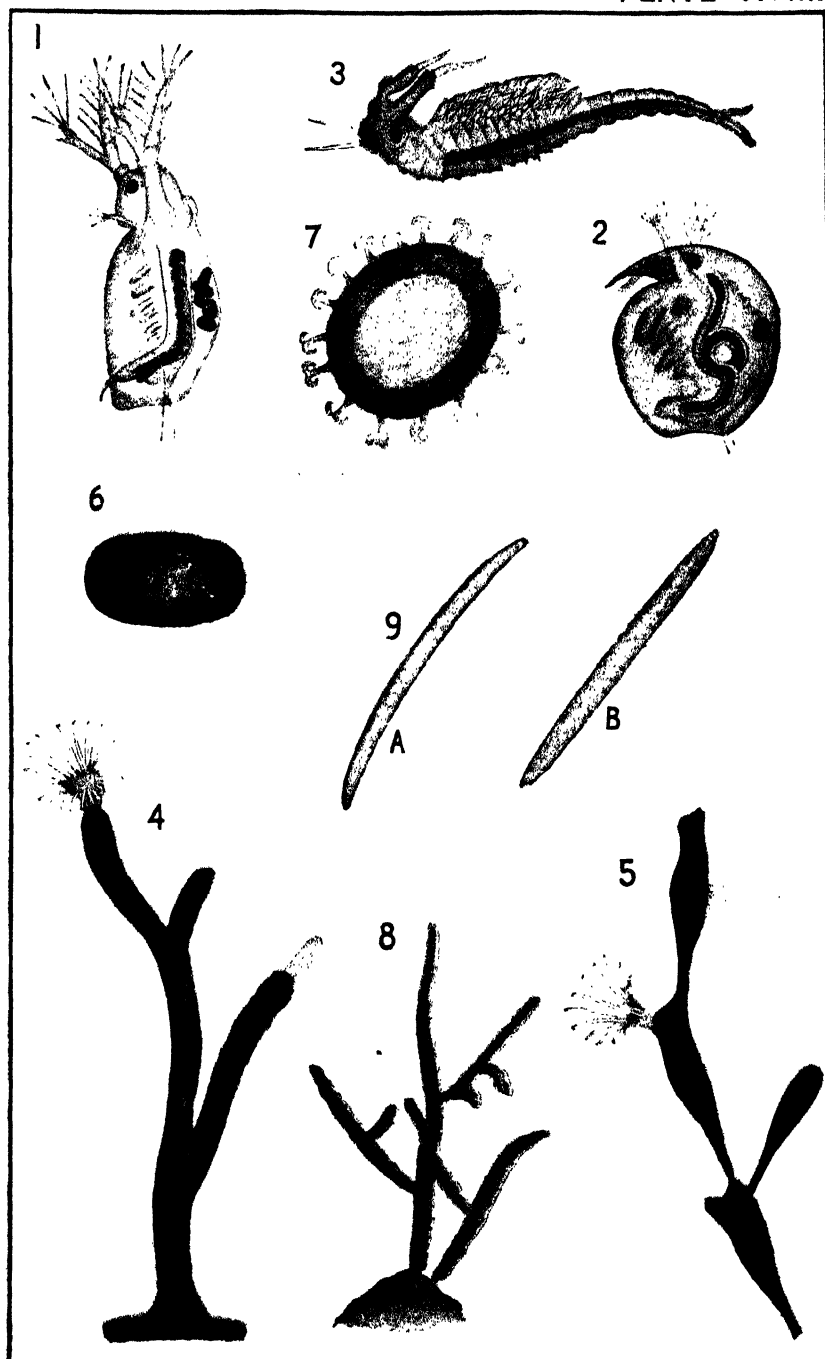


PLATE XIX
MISCELLANEOUS ORGANISMS

PLATE XIX

VERMES OR WORMS

FIGURE	PAGE
1. Anguillula. $\times 100$ (Nematoda or Round Worms).....	539
2. Nais. $\times 10$ (Oligochaeta or Bristle Worms).....	539
3. Chætonotus. $\times 250$ (Gastrotricha or Trochal Worms).....	539

ARACHNIDA OR SPIDERS AND MITES

4. Macrobiotus. $\times 250$ (Tardigrada or Water Bears).....	539
5. Hydrachna (Acarina). $\times 25$ (Hydrachnidæ or Water Mites).....	539

HYDROZOA OR HYDROIDS

6. Hydra. $\times 25$	539
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RHODOPHYCEÆ OR RED ALGÆ

7. Batrachospermum. $\times 100$	494
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CHLOROPHYCEÆ OR GREEN ALGÆ

8. Chara. $\times 75$ (Characeæ).....	480
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SPERMATOPHYTA OR SEED PLANTS

9. Elodea. $\times 1$ (Waterweed).....	539
10. Ceratophyllum. $\times 1$ (Hornwort).....	539
11. Potamogeton. $\times 1$ (Pondweed).....	539
12. Lemna. $\times 1$ (Duckweed).....	539

SCALES

